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(57) Abstract

The stability of the 20,396 dalton *Bacillus circulans* xylanase was increased by site-directed mutagenesis. Increased stability was conferred by the presence of non-native disulfide bridges, and selected N-terminal mutations. The introduction of these non-native disulfide bridges was accomplished by the examination of the three-dimensional structure of the enzyme, and choosing sites where a favorable geometry for a bridge existed. The N-terminal mutations were constructed on the basis of primary sequence comparison with other family G xylanases. The mutant proteins were examined: for their ability to retain enzymatic activity after heating, as an indication of increased thermostability; for their ability to function at elevated temperatures and for their ability to function at a more basic pH. These more stable variants are useful as an alternative to chemical bleaching of Kraft pulp in a pre-bleaching step (bio-bleaching). The pre-bleaching step involves higher temperature and pH than that normally used for these enzymes, and accordingly these variants can be advantageously used at this step. These stable xylanases are also of use in the food processing industry.

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STABLE MUTANTS OF A LOW MOLECULAR MASS XYLANASE

This application is a continuation-in-part of U.S. application 08/044,621 filed April 8, 1993.

BACKGROUND AND PRIOR ART

The present invention is directed to a modified xylanase, which shows an improved stability when compared to the naturally occurring xylanase. Specifically the present invention is directed to a modified xylanase, wherein said xylanase has increased thermostability, increased temperature optimum and/or increased pH optimum, and wherein said xylanase is modified through either the introduction of at least one non-native disulfide bridge, the introduction of at least one N-terminal mutation, or combinations thereof.

Current strategies for the production of paper use a chemical bleaching step, which is essential for the colour and quality of the paper. Chemical bleaching uses chlorine or chlorine dioxide and produces substantial amounts of by-products, which are environmental pollutants. The bleaching process can be enhanced by using an enzymatic pre-treatment with xylanase (Paice and Jurasek, 1984 Journal of Wood Chemistry and Technology, 4(2):187-198), which lowers the chlorine charge needed to affect bleaching, thereby reducing pollutants. In addition there is less bleaching chemical used, which lowers the chemical costs. New bleaching technology using, oxygen or peroxid s with xylanase, are also as effective in brightening the pulp.

The step in the process where the enzyme is applied is after a hot alkali treatment, so that the pulp is very basic and hot. Both of these conditions are sub-optimal for xylanase enzymatic activity. Many pulp mills have the capability to acidify the pulp to a pH which

is closer to the pH optimum for the enzyme and the pulp is adjusted from pH 10-11 to pH 6-8. Due to corrosion risk the pH should preferably be higher than 6.5. A xylanase with a higher pH optimum would be advantag ous, especially if the pH-adjustment step could be completely eliminated. Cooling the pulp is too energy intensive (expensive) to be used in the mill setting. Therefore, a xylanase, which is active at a higher temperature would be useful in the bio-bleaching processes. According to Nissen et al. (Xylanases for the Pulp and Paper Industry In: Xylans and Xylanases, Ed. J. Visser et al., 1992, Elsevier Science publishers B.V.) the ideal xylanase for the bleaching process would have a temperature optimum of 70°C and a pH optimum of 9.

Xylanase also has uses in non-pulp applications. Xylanases have been reported to be useful in clarifying juice and wine (Zeikus. J.G., Lee, Y.-E., and Saha, B.C. 1991. ACS Symp. Ser. 460:36-51; Beily, P. 1991. ACS Symp. Ser. 460:408-416; Woodward J. 1984. Top Enzyme Ferment. Biotechnol. 8:9-30), extracting coffee, plant oils and starch (McCleary, B.V. 1986. Int. J. Biol. Macromol. 8:349-354; Beily, P. 1991. ACS Symp. Ser. 460:408-416; Woodward J. 1984. Top Enzyme Ferment. Biotechnol. 8:9-30), for the production of food thickeners (Zeikus. J.G., Lee, Y.-E., and Saha, B.C. 1991. ACS Symp. Ser. 460:36-51), altering texture in bakery products (Maat, J., Roza. M., Verbakel, J., Stam, H., Santos da Silva, M.J., Bosse, M., Egmond, M.R., Hagemans, M.L.D., v.Gorcom, R.F.M., Hessing, J.G.M., v.d.Hodel, C.A.M.J.J., and Rotterdam,C. 1992. In Xylans and xylanases. Visser, J., Beldman, G., Kusters-van Someren, M.A. and Voragen, A.G.J., eds. Elsevier Sci pub., Amsterdam. ISBN 0-444-894-772; McCleary, B.V. 1986. Int. J. Biol. Macromol. 8:349-354), and in the washing of super precision devices and semiconductors (Takayuki, I., Shoji, S. US patent number 5078802, issue date 92-01-07). Several of these application could benefit from a thermostable xylanase, for example, food proc ssing at elevated temperatures.

A thermostable xylanase from *Thermoascus aurantiacus* was produced in US Patent 4,966,850 (Yu et al.) from a particular strain of *T. aurantiacus*, while culturing the strain at high temperature culturing conditions, however this enzyme is not a member of the family G xylanases (Gilkes et al. 1991, Microbiol. Reviews 55(2):303-315), which is the subject of this patent.

Arase et al. (FEBS 316:123-127, 1993) report improvements in thermostability of *Bacillus pumilus* xylanase through random mutagenesis of the gene by chemical mutagens. Their improvements in thermostability are minor in comparison to that of the present invention. The prior art most stable mutant maintained 40% residual activity after a short period of 20 minutes at 57°C. This mutant had a low specific activity, equivalent to 19% of the wild type *B. pumilus* xylanase. It is noted that the temperature optimum and pH optimum of these prior art mutants were not studied.

Site directed mutagenesis has been used to produce more stable proteins. Disulfide (SS) bonds in proteins restrict the degree of freedom for the unfolded state and thereby stabilize the folded state. The first type of protein stabilization performed by genetic manipulation was the introduction of disulfide bonds. One or two amino acids in the protein are replaced with cysteines; a disulfide bond forms *in vivo* or *in vitro*. If the introduced disulfide bond causes no or little tertiary structural change, the cross-links stabilizes the protein. Disulfide bonds have been engineered into T4 lysozyme (T4L) (Perry, L.J. and Wetzel, R. (1984) Science 226, 555-557; Wetzel, R., Perry, L.J. Baase, W.A. and Becktel, W.J. (1988) Proc. Nat. Acad. Sci. USA 85, 401-405), subtilisin (Wells, J.A. and Powers, D.B. (1986) J. Biol. Chem. 261,

6564-6570; Mitchinson, C. and Wells, J.A. (198), dihydrofolate reductase (DHFR) (Villafranca, J.E., Howell, E.E., Oatley, S.J., Xuong, N. and Kraut, J. (1987) Biochemistry 26, 2182-2189), and the Phage λ repressor (Cl) (Sauer, R.T., Hehir, K., Stearman, R.S. et al. (1986) Biochemistry 25, 5992-5998) with stabilization occurring in some cases but not others.

For example, in T4L the introduction of SS bonds showed an increase in thermostability of between 6 and 11°C based on reversible denaturation at pH 2 (Matsumura et al, 1989, Nature 342:291-293). This data does not show how much activity remains after heating the sample, which in a functional sense is what is important for an industrial enzyme. The RNASE H SS bond mutant is also stabilized by 11.8°C as measured by reversible thermal denaturation, but has no enzymatic activity (Kanaya et al, 1991, Journal of Biological Chemistry 226(10):6038-6044). In DHFR the artificial SS bond contributes to stability of the protein against chemical denaturation, but does not confer thermostability (Villafranca et al, 1987, Biochemistry 26:2182-2189). A similar situation occurs with subtilisin, where 5 different engineered SS bonds do not confer thermostability (Mitchinson and Wells, 1989, Biochemistry 28:4807-4815).

SUMMARY OF INVENTION

According to the present invention there is provided a modified xylanase, wherein said xylanase has increased stability and wherein said xylanase is modified through either the introduction of at least one non-native disulfide bridge, the introduction of at least one N-terminal mutation, or combinations thereof. The modified xylanases of the present invention exhibit at least one of the following traits: increased thermostability, increased temperature optimum or increased pH optimum.

In one embodiment of the present invention, the modified xylanase has been modified by the introduction of an intra-molecular disulfide bridge between a cysteine amino acid, which has been introduced on the last strand of sheet III, and a cysteine amino acid, which has been introduced on the alpha helix.

In a further embodiment of the present invention, the modified xylanase has been modified by the introduction of an inter-molecular disulfide bridge between two xylanase molecules, wherein a cysteine amino acid has been introduced in each of said two molecules.

In a further embodiment of the present invention, the modified xylanase has been modified by the introduction of a mutations at the N-terminus of the xylanase.

In one embodiment of the present invention there is provided a modified family G xylanase essentially having the structure of the *B. circulans* enzyme or mutated to essentially have this structure, wherein said xylanase has increased stability and wherein said xylanase is modified through either the introduction of at least one non-native disulfide bridge, the introduction of at least one N-terminal mutation, or combinations thereof;

wherein the disulfide bridge is an intra-molecular bridge between a cysteine amino acid, which has been introduced on the last strand of beta-sheet III, and a cysteine amino acid, which has been introduced on the alpha helix, or on either side, adjacent to the alpha helix or the disulphide bridge is an inter-molecular bridge between two xylanase molecules, wherein a cysteine amino acid has been introduced in each of said two molecules, on the external region; and

wherein the N-terminal mutation is selected from at least one mutation of the group consisting of the introduction of tyrosine at amino acid position 8,

introduction of phenylalanine at amino acid position 8, introduction of proline at amino acid position 22, and introduction of an N-terminal to C-terminal disulfide bridge, and wherein these N-terminal mutations can be used in combination with other N-terminal mutation introduced at amino acid position 1 to 25 of the N-terminal region, based on the amino acid numbering from *B. circulans* xylanase.

In a further embodiment of the present invention there is provided a modified family G xylanase essentially having the structure of the *B. circulans* enzyme or mutat d to essentially have this structure, wherein said xylanase has increased stability and wherein said xylanase is modified through either the introduction of at least one non-native disulfide bridge, the introduction of at least one N-terminal mutation, or combinations thereof; and wherein said modified xylanase is produced from clones selected from the group consisting of TS1, TS2, TS3, TS3a, TS5a, TS6a, TS7a, TS10a, TS14a, TS15a, TS17a, TS19a, TS20a and TS21a.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows the multiple sequence alignment among low molecular weight xylanases.

Figure 2 shows a stereodiagram of the overall fold of the three-dimensional crystal structure of the *Bacillus circulans* xylanase. The strands of beta-sheet are represented by arrows and the alpha-helix is represented by a cylinder. The three beta-sheets referred to in the text are indicated by I, II, and III.

Figure 3 shows a stereodiagram of the superimposed three-dimensional crystal structures of the xylanases from *B. circulans* and *T. harzianum*. The structures are drawn such that the positions of the alpha-carbons of each

amino acid are linked by a straight line. The *B. circulans* structure is drawn in a thick line and the *T. harzianum* structure is drawn in a thin line. The *T. harzianum* xylanase has a slightly more open active site due to different intermolecular crystal contacts.

Figure 4 shows a stereodiagram of the superimposed structures of the wild-type and disulfide-containing mutant (TS1) of the *B. circulans* xylanase in the vicinity of the mutation (residues 100 and 148). The wild-type enzyme is drawn in thick lines and the mutant is drawn in thin lines.

Figure 5 shows a stereodiagram of the superimposed structures of the xylanases from *B. circulans* and *T. harzianum* in the vicinity of the disulfide bond shown in Figure 4. The *B. circulans* structure is drawn in a thick line and the *T. harzianum* structure is drawn in a thin line.

Figure 6 shows the native sequence (SEQ ID NO:1) of the *B. subtilis* xylanase gene from plasmid pBSX. The binding sites for the PCR primers and the S100C mutagenic primer are underlined.

Figure 7 shows the semi-synthetic gene sequence (SEQ ID NO:2) for *B. circulans* xylanases. The sites for UDNA mutagenesis to produce TS2 are shown with underlining.

Figure 8 shows the SDS PAGE results of monomer and dimer fraction from TS4a, TS4M and TS4D. Lane 1 is TS4a monomer fraction, reduced; Lane 2 is as in lane 1 but non-reduced; Lane 3 is TS4a dimer fraction, reduced; Lane 4 is as lane 3 but non-reduced; Lane 5 is TS4M monomer fraction, reduced; Lane 6 is as in lane 5 but non-reduced; Lane 7 is TS4D dimer fraction reduced; Lane 8 is as lane 7 but non-reduced; Lane 9 is BCX wild type reduced; Lane 10

is as in lane 9 but non-reduced. For lanes 1-4, approximately 1 μg of protein was loaded and for the other lanes, approximately 2 μg was loaded.

Figure 9 shows the complete synthetic gene sequence (SEQ ID NO:3) encoding the *B. circulans* xylanase in the plasmid pXYbc.

Figure 10 shows the electrophoretic mobility of the disulfide bridge containing mutants of *B. circulans* xylanase. Lane 1 shows the molecular weight standard, lane 2 is the BCX wild-type, reduced, lane 3 is as in lane 2 but non-reduced, lane 4 is the TS1 mutant, reduced, lane 5 is the TS1 mutant, non-reduced, lane 6 is the TS2 mutant, reduced and lane 7 is a TS2 mutant, non-reduced.

Figure 11 shows a comparison of the thermostability at 58°C of various mutants of *B. circulans* xylanase. The curve shown for TS4 is for the mixture of both monomer and dimer.

Figure 12 shows a comparison of the thermostability at 61°C of various mutants of the *B. circulans* xylanases. The curve shown for TS4 is for the mixture of both monomer and dimer.

Figure 13 shows the thermostability of the TS4a (S179C) dimer at 58°C.

Figure 14 shows the thermostability at 58°C of *B. circulans* xylanase mutants TS3 and TS4D, which are combinations of mutants shown in Figures 11 and 12.

Figure 15 shows the thermostability at 62°C of *B. circulans* xylanase mutants TS3 and TS4D, which are combinations of mutants shown in Figures 11 and 12.

Figure 16 shows the thermostability at 64°C of *B. circulans* xylanase mutants TS3 and TS4D, which are combinations of mutations shown in Figures 11 and 12.

Figure 17 illustrates the effect of temperature on enzyme activity. With the thermostable xylanase mutants shown, an increase of 18°C resulted in between 2.6 and 4 fold increase in activity.

Figure 18 shows the thermostability of xylanase BCX and mutants wherein xylanase from the wild-type or mutant strains were heated at various temperatures for 30 minutes. After cooling to 20°C a residual enzymatic activity of the heated samples were determined via the HBAH assay at 40°C.

Figure 19 shows the molecular structure of residue 6-10 and 16-20 of the N-terminus of the wild-type BCX. CA represents the alpha carbon atom of each residue. CB is the beta-carbon atom of the side-chain. OD1 is the delta oxygen atom in the side-chain of asparagine-8. The number next to CA, CB and OD1 designates the residue to which these atoms belong. The two asterisks are the two buried water molecules. The broken lines represent the hydrogen bonds.

Figure 20 shows the molecular structure of the same region as Figure 19, illustrating the postulated effect of the asparagine to tyrosine mutation at residue-8 (TS5a).

Figure 21 shows the average main-chain B-factors for the xylanase from *B. circulans*. The B-factor is a measure of the spreading out of electron density. It is thus a measure of the mobility of the atoms in a structure. This plot shows the B-factors (or mobility) averaged over all main-chain atoms in each amino acid. Thus the regions near

the amino-terminus and near residue 120 are the most mobile parts of the structure.

Figure 22 shows the average main-chain B-factors for the xylanase from *T. harzianum*.

Figure 23 shows the average main-chain B-factors for the disulfide-containing mutant (TS1) of the xylanase from *B. circulans*.

Figure 24 shows a Ramachandran plot for the xylanase from *B. circulans*. The two residues (D121 and A165) with main-chain dihedral angles outside of normal limits are labelled. Glycines are shown with circles, while all other amino acids are shown with "plus" signs.

Figure 25 shows a Ramachandran plot for the xylanase from *T. harzianum*. The three residues (T2, N11 and S181) with main-chain dihedral angles outside of normal limits are labelled. Glycines are shown with circles, while all other amino acids are shown with "plus" signs.

Figure 26 shows enzymatic activity of selected xylanase mutants in a thirty minute assay on soluble xylan at temperatures ranging from 40°C to 80°C.

Figure 27 shows the enzymatic activity of selected xylanase mutants at a pH ranging from 4 to 10.

Figure 28 demonstrates the bleaching of pulp at pH 8 by selected xylanase mutants at temperatures ranging from 40°C to 70°C.

DETAILED DESCRIPTION OF THE INVENTION

The present invention is related to modified xylanases belonging to the family G xylanases (Henrissat

and Bairoch, Biochem, J. 293: 781-788, 1993 and Gebler et al., J. Biol. Chem. 266: 12559-12561, 1992; incorporated herewith by reference) recently renamed to family 11 (Henrissat and Bairoch, Biochem. J. 293: 7681-788, 1993). The original family G classification was based on the primary sequence comparison and hydrophobic cluster analysis of 4 protein sequences that were in the database at the time the paper, regarding the original classification, was published. Since that time, an additional 16 sequences have been added to this group. In addition to the sequence comparisons, the group definition, also included the stereochemical outcome of the hydrolysis reaction (catalytic mechanism). The grouping by sequence analysis predicts the protein folding will be similar, and the examination of the stereochemistry of the reaction is an indirect measure of how the active site residues are oriented (the prediction is enzymes with similar structure give the same stereochemistry in the reaction products).

The complete amino acid sequence of the low molecular weight xylanase has been determined from a number of different bacterial and fungal sources (M. Yaguchi et al., Amino Acid Sequence of the Low-Molecular-Weight Xylanase from *Trichoderma viride*, Xylans and Xylanases, edited by J. Visser et al, 1992, 149-154). The sequence comparison reported in the reference referred above has been extended to show the sequence similarities and differences between the bacteria and fungal family G xylanases as listed below.

BACTERIAL

Bacillus pumilus

Fukusaki, E., Panbangred, W., Shinmyo, A., Okada, H. FEBS Letters 171:197-201 (1984)

Clostridium acetobutylicum, XYN B

Zappe, H., Jones, W.A., Woods, D.R. Nucleic Acids Research 18:2179 (1990)

Ruminococcus flavefaciens

Zhang, J., Flint, H.J. EMBL database accession number Z11127 (1992)

Streptomyces sp. No. 36a

Nagashima, M., Okumoto, Y., Okanishi, M. Trends in Actinomycetoligia, 91-96 (1989)

Streptomyces lividans, XYN B and XYN C

Shareck, F., Roy, C., Yaguchi, M., Morosoli, R., Kluepfel, D.Gene, 107:75-82 (1991)

Bacillus circulans

Yang, R.C.A., MacKenzie, C.R., Narang, R.A. Nucleic Acids Res. 16:7187 (1988)

Bacillus subtilis

Paice, M.G., Bourbonnais, R., Desrochers, M., Jurasek, L., Yaguchi, M. Arch. Microbiol. 144:201-206 (1986)

FUNGAL

Trichoderma reesei, XYN I and XYN II

Torronene, A., Mach, R.L., Messner, R., Gonzalez, R., Kalkkinen, N., Harkki, A., Kubicek, C.P. Bio/Technology 10:1461-1465 (1992)

Trichoderma viride, 20KD

Yaguchi, M., Roy, C., Ujlie, M., Watson, D.C., Wakarchuk, W. Xylans and Xylanases, ed. by J. Visser et al., Elsevier, pp. 149-154 (1992)

Trichoderma harzianum, 20KD

Yaguchi, M., Roy, C., Watson, D.C., Rollin, F., Tan. L.U.L., Senior, D.J., Saddler, J.N. Xylans and Xylanases, ed. by J. Visser et al., Elsevier, pp. 435-438 (1992)

Schizophyllum commune, Xylanase A

Oku, T., Roy, C., Watson, D.C., Wakarchuk, W., Yaguchi, M., Jurasek, L., Paice, M.G. (unpublished)

Aspergillus niger var. awamori

Maat, J., Roza, M., Verbakel, J., Stam, H., Santos da Silva, M.J., Egmond, M.R., Hagemans, M.L.D., Gorcom, R.F.M.v., Hessing, J.G.M., Hondel, C.A.M.J.J.v.d., Rotterdam, C.v. Xylans and Xylanases, ed. by J. Visser et al., Elsevier, pp. 349-360 (1992)

Aspergillus tubigensis, XYL A

de Graaff, L.H., van den Broeck, H.C., van Ooijan, A.J.J., Visser, J. Xylans and Xylanases, ed. by J. Visser et al., Elsevier, pp. 235-246 (1992)

Figure 1 shows a multiple sequence alignment among low molecular mass xylanases obtained with GeneWorks version 2.2.1 (IntelliGenetics, Inc., Mountain View, CA), which has been manually edited to conform to the structural homology between the xylanases from *B. circulans* and *T. harzianum*. The extended N-terminal sequence of the *C. acetobutylicum* xylanase B (residues 1-31) and the C-terminal sequences of *S. lividans*, xylanase B (residues 216-293) are not shown.

Table 1 shows the amino acid sequence identity (percentage) between different groups of family G xylanases, when compared to *B pumilus*, *B. circulans*, *T. harzianum* and *Schizo. commune* as references. Due to the sequence homology among related xylanases, one could expect that mutations for the introduction of SS bonds or for the introduction of mutations at the N-terminal end of the xylanase, as demonstrated in the examples below for *B. circulans* xylanase, can be extended to other xylanases from bacterial or fungal sources to produce similar effects.

Table 1

Amino Acid Sequence Identity (%) between the Family G Xylanases

	Bacillus pumilus	*Bacillus circulans*	*Trichoderma harzianum*	*Schizo. commune*
Bacterial				
B. pumilus	-	43	46	40
C. aceto-butylicum	71	42	42	41
R. flavefaciens	46	36	41	39
Strept. sp. 36a	49	57	50	47
S. lividans, B	50	56	51	47
S. lividans, C	49	60	52	51
B. circulans	43	-	51	50
B. subtilis	43	99	51	50
Fungal				
T. reesei, II	47	51	95	53
T. viride	48	50	94	53
T. harzianum	46	51	-	54
S. commune	40	50	54	-
A. niger awamori	34	42	41	38
A. tubigensis, A	34	43	41	37
T. reesei, I	37	46	50	38

The xylanase from *B. circulans* is functionally identical to that from *B. subtilis*, which will be seen later in

the Examples. These proteins differ at only one residue and are among the shortest xylanases, with their N-terminus lacking at least 10 amino acid residues compared to other xylanases.

The three-dimensional crystal structure of the *B. circulans* xylanase is represented in Figure 2. This ribbon representation shows that the structure is composed of three beta-sheets and one alpha-helix. The first two beta-sheets are roughly parallel, while sheet III is at about a 90° angle to sheet II. Sheets I and II are each composed of five strands, while sheet III contains 6 strands. The alpha-helix lies across the back of sheet III and the last two strands of sheet III fold over one edge of the alpha-helix. The active site lies in the cleft between sheets II and III. The x-ray co-ordinates of the structure is given below.

ATOM	1	CB	ALA	1	32.404	42.580	22.419	1.00 19.66
ATOM	2	C	ALA	1	34.898	42.615	22.344	1.00 20.06
ATOM	3	O	ALA	1	35.540	41.613	22.023	1.00 20.55
ATOM	6	N	ALA	1	33.610	42.325	20.269	1.00 18.68
ATOM	8	CA	ALA	1	33.627	42.997	21.599	1.00 19.67
ATOM	9	N	SER	2	35.283	43.440	23.308	1.00 20.15
ATOM	11	CA	SER	2	36.455	43.158	24.114	1.00 20.60
ATOM	12	CB	SER	2	37.000	44.436	24.753	1.00 22.52
ATOM	13	OG	SER	2	37.614	45.262	23.783	1.00 25.52
ATOM	15	C	SER	2	36.062	42.184	25.210	1.00 20.08
ATOM	16	O	SER	2	34.967	42.270	25.786	1.00 19.93
ATOM	17	N	THR	3	36.930	41.215	25.440	1.00 18.98
ATOM	19	CA	THR	3	36.715	40.235	26.475	1.00 18.01
ATOM	20	CB	THR	3	37.469	38.943	26.145	1.00 18.17
ATOM	21	OG1	THR	3	38.849	39.247	25.891	1.00 17.58

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ATOM     23  CG2 THR   3      36.860  38.273  24.920
1.00 17.34
ATOM     24  C   THR   3      37.306  40.884  27.713
1.00 17.67
ATOM     25  O   THR   3      38.232  41.694  27.610
1.00 18.99
ATOM     26  N   ASP   4      36.739  40.601  28.874
1.00 16.43
ATOM     28  CA  ASP   4      37.252  41.190  30.104
1.00 16.02
ATOM     29  CB  ASP   4      36.284  42.252  30.644
1.00 16.80
ATOM     30  CG  ASP   4      34.886  41.706  30.907
1.00 19.10
ATOM     31  OD1 ASP   4      34.741  40.512  31.221
1.00 17.29
ATOM     32  OD2 ASP   4      33.923  42.495  30.810
1.00 21.94
ATOM     33  C   ASP   4      37.506  40.143  31.161
1.00 15.35
ATOM     34  O   ASP   4      37.721  40.470  32.325
1.00 16.73
ATOM     35  N   TYR   5      37.455  38.875  30.769
1.00 13.80
ATOM     37  CA  TYR   5      37.665  37.812  31.722
1.00 12.53
ATOM     38  CB  TYR   5      36.318  37.186  32.119
1.00 12.90
ATOM     39  CG  TYR   5      36.430  36.223  33.270
1.00 14.19
ATOM     40  CD1 TYR   5      36.583  36.679  34.577
1.00 15.98
ATOM     41  CE1 TYR   5      36.748  35.786  35.632
1.00 17.93
ATOM     42  CD2 TYR   5      36.438  34.855  33.052
1.00 14.54
ATOM     43  CE2 TYR   5      36.603  33.966  34.092
1.00 16.97
ATOM     44  CZ  TYR   5      36.762  34.431  35.379
1.00 17.23
ATOM     45  OH  TYR   5      36.972  33.528  36.401
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ATOM     47  C   TYR   5      38.610  36.754  31.175
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ATOM     49  N   TRP   6      39.523  36.309  32.027
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ATOM     51  CA  TRP   6      40.498  35.270  31.691
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ATOM     52  CB  TRP   6      41.922  35.830  31.788
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ATOM     53  CG  TRP   6      43.050  34.813  31.806
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ATOM     54  CD2 TRP   6      43.420  33.895  30.761
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ATOM      56  CE3 TRP     6      42.879  33.570  29.515
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ATOM      57  CD1 TRP     6      43.982  34.661  32.791
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ATOM      58  NE1 TRP     6      44.912  33.718  32.425
1.00 14.48
ATOM      60  CZ2 TRP     6      45.236  32.271  30.404
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ATOM      61  CZ3 TRP     6      43.518  32.607  28.740
1.00 13.00
ATOM      62  CH2 TRP     6      44.681  31.974  29.187
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ATOM      63  C   TRP     6      40.292  34.122  32.670
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ATOM      64  O   TRP     6      40.399  34.289  33.890
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ATOM      67  CA  GLN     7      39.656  31.763  32.877
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ATOM      68  CB  GLN     7      38.380  31.105  32.332
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ATOM      69  CG  GLN     7      38.105  29.698  32.821
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ATOM      70  CD  GLN     7      37.879  29.624  34.314
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ATOM      71  OE1 GLN     7      37.712  30.644  34.993
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ATOM      72  NE2 GLN     7      37.867  28.411  34.838
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ATOM      75  C   GLN     7      40.843  30.818  32.726
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ATOM      76  O   GLN     7      41.211  30.455  31.612
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ATOM      79  CA  ASN     8      42.615  29.550  33.784
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ATOM      80  CB  ASN     8      43.924  30.351  33.687
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ATOM      81  CG  ASN     8      45.166  29.462  33.674
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ATOM      82  OD1 ASN     8      46.279  29.935  33.916
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ATOM      83  ND2 ASN     8      44.990  28.192  33.377
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ATOM      86  C   ASN     8      42.604  28.712  35.045
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ATOM      87  O   ASN     8      43.215  29.075  36.057
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ATOM      90  CA  TRP     9      41.766  26.703  36.113
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ATOM      92  CG  TRP     9      40.058  25.646  37.680
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ATOM      93  CD2 TRP     9      39.703  24.260  37.548
1.00 19.90
ATOM      94  CE2 TRP     9      39.600  23.728  38.851
1.00 20.63
ATOM      95  CE3 TRP     9      39.462  23.420  36.455
1.00 20.29
ATOM      96  CD1 TRP     9      40.151  25.888  39.025
1.00 20.86
ATOM      97  NE1 TRP     9      39.879  24.740  39.731
1.00 21.73
ATOM      99  CZ2 TRP     9      39.268  22.397  39.090
1.00 20.83
ATOM     100  CZ3 TRP     9      39.133  22.100  36.694
1.00 20.61
ATOM     101  CH2 TRP     9      39.038  21.600  38.003
1.00 20.23
ATOM     102  C   TRP     9      42.256  25.300  35.799
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ATOM     103  O   TRP     9      41.974  24.757  34.732
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ATOM     104  N   THR    10      42.978  24.714  36.745
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ATOM     106  CA  THR    10      43.461  23.352  36.609
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ATOM     107  CB  THR    10      44.929  23.291  36.133
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ATOM     108  OG1 THR    10      45.385  21.934  36.177
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ATOM     110  CG2 THR    10      45.831  24.146  37.021
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ATOM     111  C   THR    10      43.362  22.696  37.976
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ATOM     112  O   THR    10      43.419  23.378  39.000
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ATOM     113  N   ASP    11      43.145  21.387  38.000
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ATOM     115  CA  ASP    11      43.101  20.686  39.277
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ATOM     116  CB  ASP    11      42.200  19.448  39.228
1.00 16.43
ATOM     117  CG  ASP    11      42.629  18.417  38.186
1.00 16.04
ATOM     118  OD1 ASP    11      43.738  18.498  37.612
1.00 16.20
ATOM     119  OD2 ASP    11      41.819  17.506  37.945
1.00 17.65
ATOM     120  C   ASP    11      44.529  20.336  39.724
1.00 17.44
ATOM     121  O   ASP    11      44.723  19.661  40.731
1.00 18.70
ATOM     122  N   GLY    12      45.517  20.780  38.946
1.00 17.45
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ATOM	124	CA	GLY	12	46.913	20.547	39.287
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ATOM	125	C	GLY	12	47.616	19.394	38.593
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ATOM	126	O	GLY	12	48.841	19.251	38.699
1.00	18.34						
ATOM	127	N	GLY	13	46.862	18.568	37.881
1.00	16.38						
ATOM	129	CA	GLY	13	47.466	17.443	37.202
1.00	15.25						
ATOM	130	C	GLY	13	48.141	17.771	35.887
1.00	15.48						
ATOM	131	O	GLY	13	47.608	18.530	35.070
1.00	15.97						
ATOM	132	N	GLY	14	49.319	17.187	35.687
1.00	14.01						
ATOM	134	CA	GLY	14	50.062	17.366	34.456
1.00	13.22						
ATOM	135	C	GLY	14	50.476	18.783	34.128
1.00	13.48						
ATOM	136	O	GLY	14	50.581	19.642	35.009
1.00	15.09						
ATOM	137	N	ILE	15	50.707	19.026	32.845
1.00	12.64						
ATOM	139	CA	ILE	15	51.145	20.327	32.387
1.00	13.51						
ATOM	140	CB	ILE	15	52.415	20.196	31.469
1.00	16.01						
ATOM	141	CG2	ILE	15	52.750	21.523	30.777
1.00	16.26						
ATOM	142	CG1	ILE	15	53.615	19.720	32.291
1.00	17.95						
ATOM	143	CD1	ILE	15	53.595	18.263	32.618
1.00	22.36						
ATOM	144	C	ILE	15	50.054	21.032	31.604
1.00	11.40						
ATOM	145	O	ILE	15	49.447	20.438	30.715
1.00	11.13						
ATOM	146	N	VAL	16	49.780	22.276	31.984
1.00	11.65						
ATOM	148	CA	VAL	16	48.811	23.124	31.291
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ATOM	149	CB	VAL	16	47.501	23.332	32.103
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ATOM	150	CG1	VAL	16	46.539	24.263	31.332
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ATOM	151	CG2	VAL	16	46.833	21.989	32.368
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ATOM	152	C	VAL	16	49.515	24.464	31.084
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ATOM	153	O	VAL	16	49.699	25.225	32.028
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ATOM    158  CG  ASN  17      52.558  26.962  28.153
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ATOM    159  OD1 ASN  17      52.808  27.102  26.954
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ATOM    160  ND2 ASN  17      52.840  27.888  29.049
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ATOM    163  C   ASN  17      49.674  26.840  28.871
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ATOM    164  O   ASN  17      49.446  26.720  27.663
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ATOM    165  N   ALA  18      49.038  27.706  29.654
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ATOM    167  CA  ALA  18      48.014  28.616  29.143
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ATOM    168  CB  ALA  18      46.831  28.682  30.114
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ATOM    169  C   ALA  18      48.601  29.999  28.941
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ATOM    170  O   ALA  18      49.201  30.566  29.856
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ATOM    171  N   VAL  19      48.415  30.551  27.751
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ATOM    173  CA  VAL  19      48.925  31.871  27.429
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ATOM    174  CB  VAL  19      49.800  31.839  26.167
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ATOM    175  CG1 VAL  19      50.307  33.240  25.850
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ATOM    176  CG2 VAL  19      50.964  30.862  26.357
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ATOM    177  C   VAL  19      47.790  32.865  27.207
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ATOM    178  O   VAL  19      46.901  32.643  26.384
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ATOM    182  CB  ASN  20      46.747  35.697  29.247
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ATOM    183  CG  ASN  20      45.715  36.812  29.294
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ATOM    185  ND2 ASN  20      45.390  37.255  30.501
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ATOM    190  N   GLY  21      46.941  35.814  25.571
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ATOM    195  N   SER  22     47.115  38.719  23.271
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ATOM    198  CB  SER  22     47.327  40.815  21.970
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ATOM    199  OG  SER  22     47.248  40.141  20.732
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ATOM    201  C   SER  22     45.083  39.974  22.692
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ATOM    202  O   SER  22     44.634  39.007  22.060
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ATOM    205  CA  GLY  23     42.925  41.053  22.791
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ATOM    206  C   GLY  23     42.198  39.826  23.305
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ATOM    207  O   GLY  23     42.338  39.444  24.461
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ATOM    208  N   GLY  24     41.429  39.199  22.425
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ATOM    210  CA  GLY  24     40.682  38.019  22.812
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ATOM    211  C   GLY  24     41.371  36.717  22.458
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ATOM    216  CB  ASN  25     44.532  35.893  20.833
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ATOM    218  OD1 ASN  25     46.499  34.512  20.908
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ATOM    219  ND2 ASN  25     44.790  33.823  19.620
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ATOM    222  C   ASN  25     44.048  34.839  23.033
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ATOM    226  CA  TYR  26     44.696  32.715  24.016
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ATOM    227  CB  TYR  26     43.802  32.457  25.243
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ATOM    228  CG  TYR  26     42.670  31.472  25.023
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ATOM    231  CD2 TYR  26      42.851  30.108  25.240
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ATOM    232  CE2 TYR  26      41.815  29.210  25.057
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ATOM    233  CZ  TYR  26      40.577  29.682  24.652
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ATOM    234  OH  TYR  26      39.534  28.794  24.438
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ATOM    236  C   TYR  26      45.122  31.403  23.385
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ATOM    238  N   SER  27      46.087  30.735  24.007
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ATOM    240  CA  SER  27      46.528  29.440  23.516
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ATOM    241  CB  SER  27      47.759  29.566  22.604
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ATOM    242  OG  SER  27      48.833  30.187  23.275
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ATOM    245  O   SER  27      46.985  29.013  25.825
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ATOM    246  N   VAL  28      46.728  27.245  24.471
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ATOM    249  CB  VAL  28      45.692  25.668  26.146
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ATOM    250  CG1 VAL  28      46.023  24.931  27.454
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ATOM    251  CG2 VAL  28      44.641  26.726  26.377
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ATOM    252  C   VAL  28      47.724  25.086  24.929
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ATOM    257  CB  ASN  29      50.977  23.683  25.060
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ATOM    258  CG  ASN  29      51.225  24.717  24.004
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ATOM    259  OD1 ASN  29      50.951  24.496  22.820
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ATOM    260  ND2 ASN  29      51.718  25.868  24.420
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ATOM    267  CA  TRP  30      48.589  20.575  27.619  1.00  7.76
ATOM    268  CB  TRP  30      47.115  20.598  28.066  1.00  7.93
ATOM    269  CG  TRP  30      46.123  19.856  27.209  1.00  7.36
ATOM    270  CD2 TRP  30      45.383  20.374  26.090  1.00  7.38
ATOM    271  CE2 TRP  30      44.448  19.387  25.719  1.00  6.82
ATOM    272  CE3 TRP  30      45.411  21.580  25.379  1.00  8.62
ATOM    273  CD1 TRP  30      45.632  18.610  27.444  1.00  6.99
ATOM    274  NE1 TRP  30      44.618  18.321  26.562  1.00  7.92
ATOM    276  CZ2 TRP  30      43.542  19.568  24.657  1.00  9.07
ATOM    277  CZ3 TRP  30      44.510  21.759  24.330  1.00  7.97
ATOM    278  CH2 TRP  30      43.592  20.757  23.982  1.00  7.80
ATOM    279  C   TRP  30      49.112  19.166  27.431  1.00  7.73
ATOM    280  O   TRP  30      49.151  18.641  26.305  1.00  8.50
ATOM    281  N   SER  31      49.532  18.553  28.530  1.00  8.50
ATOM    283  CA  SER  31      50.095  17.218  28.466  1.00  9.39
ATOM    284  CB  SER  31      51.615  17.333  28.268  1.00 11.08
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ATOM    291  CA  ASN  32      48.934  14.290  30.648  1.00  9.24
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ATOM    293  CG  ASN  32      50.856  12.828  29.931  1.00 13.54
ATOM    294  OD1 ASN  32      50.311  11.835  29.448  1.00 15.18
ATOM    295  ND2 ASN  32      52.010  13.300  29.487  1.00 14.28
ATOM    298  C   ASN  32      48.306  14.979  31.844  1.00  9.76
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ATOM    303  CB  THR  33      45.734  17.580  31.943
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ATOM    304  OG1 THR  33      44.705  17.033  31.099
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ATOM    306  CG2 THR  33      46.668  18.456  31.117
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ATOM    307  C   THR  33      45.471  15.679  33.450
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ATOM    308  O   THR  33      45.203  14.490  33.268
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ATOM    311  CA  GLY  34      43.804  15.992  35.187
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ATOM    312  C   GLY  34      42.716  16.743  34.416
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ATOM    313  O   GLY  34      42.654  16.646  33.183
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ATOM    314  N   ASN  35      41.924  17.563  35.098
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ATOM    318  CG  ASN  35      38.361  18.897  34.531
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ATOM    320  ND2 ASN  35      38.301  18.905  33.204
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ATOM    323  C   ASN  35      41.290  19.823  34.496
1.00  9.12
ATOM    324  O   ASN  35      41.876  20.282  35.476
1.00 10.17
ATOM    325  N   PHE  36      41.061  20.545  33.406
1.00  8.58
ATOM    327  CA  PHE  36      41.396  21.966  33.357
1.00  7.71
ATOM    328  CB  PHE  36      42.832  22.193  32.856
1.00  9.56
ATOM    329  CG  PHE  36      43.018  21.898  31.389
1.00  9.71
ATOM    330  CD1 PHE  36      42.835  22.898  30.434
1.00  9.80
ATOM    331  CD2 PHE  36      43.322  20.618  30.959
1.00  9.86
ATOM    332  CE1 PHE  36      42.947  22.618  29.082
1.00 10.32
ATOM    333  CE2 PHE  36      43.437  20.335  29.600
1.00 11.13
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ATOM	334	CZ	PHE	36	43.247	21.337	28.667	1.00 10.13
ATOM	335	C	PHE	36	40.433	22.652	32.407	1.00 7.24
ATOM	336	O	PHE	36	39.856	22.003	31.528	1.00 8.20
ATOM	337	N	VAL	37	40.269	23.956	32.589	1.00 7.92
ATOM	339	CA	VAL	37	39.405	24.776	31.739	1.00 8.27
ATOM	340	CB	VAL	37	38.007	25.044	32.359	1.00 8.91
ATOM	341	CG1	VAL	37	37.165	25.888	31.382	1.00 10.27
ATOM	342	CG2	VAL	37	37.300	23.735	32.679	1.00 10.16
ATOM	343	C	VAL	37	40.109	26.109	31.568	1.00 8.48
ATOM	344	O	VAL	37	40.426	26.792	32.556	1.00 9.26
ATOM	345	N	VAL	38	40.356	26.474	30.315	1.00 7.80
ATOM	347	CA	VAL	38	41.027	27.727	29.988	1.00 8.41
ATOM	348	CB	VAL	38	42.445	27.453	29.421	1.00 9.47
ATOM	349	CG1	VAL	38	43.098	28.748	28.947	1.00 11.38
ATOM	350	CG2	VAL	38	43.305	26.766	30.470	1.00 10.26
ATOM	351	C	VAL	38	40.217	28.451	28.921	1.00 8.22
ATOM	352	O	VAL	38	39.717	27.825	28.002	1.00 9.27
ATOM	353	N	GLY	39	40.091	29.763	29.029	1.00 8.83
ATOM	355	CA	GLY	39	39.344	30.479	28.013	1.00 8.67
ATOM	356	C	GLY	39	39.214	31.960	28.280	1.00 9.03
ATOM	357	O	GLY	39	39.600	32.449	29.348	1.00 10.05
ATOM	358	N	LYS	40	38.703	32.676	27.287	1.00 8.28
ATOM	360	CA	LYS	40	38.493	34.109	27.411	1.00 7.76
ATOM	361	CB	LYS	40	39.372	34.892	26.432	1.00 8.70
ATOM	362	CG	LYS	40	40.787	35.076	26.926	1.00 10.94
ATOM	363	CD	LYS	40	41.525	36.071	26.066	1.00 10.75
ATOM	364	CE	LYS	40	42.861	36.392	26.700	1.00 11.72
ATOM	365	NZ	LYS	40	43.653	37.312	25.843	1.00 10.91

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ATOM    369  C   LYS  40      37.033  34.432  27.172
1.00  7.68
ATOM    370  O   LYS  40      36.346  33.740  26.418
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ATOM    371  N   GLY  41      36.567  35.485  27.824
1.00  7.96
ATOM    373  CA  GLY  41      35.189  35.866  27.666
1.00  8.17
ATOM    374  C   GLY  41      34.807  37.051  28.510
1.00  8.18
ATOM    375  O   GLY  41      35.547  38.042  28.593
1.00  8.33
ATOM    376  N   TRP  42      33.665  36.916  29.174
1.00  8.49
ATOM    378  CA  TRP  42      33.093  37.988  29.977
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ATOM    379  CB  TRP  42      31.790  38.455  29.304
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ATOM    380  CG  TRP  42      32.066  38.913  27.902
1.00 10.97
ATOM    381  CD2 TRP  42      32.159  38.091  26.725
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ATOM    382  CE2 TRP  42      32.562  38.926  25.667
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ATOM    383  CE3 TRP  42      31.948  36.730  26.469
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ATOM    384  CD1 TRP  42      32.396  40.174  27.512
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ATOM    385  NE1 TRP  42      32.702  40.191  26.176
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ATOM    387  CZ2 TRP  42      32.761  38.447  24.369
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ATOM    388  CZ3 TRP  42      32.147  36.254  25.178
1.00 12.72
ATOM    389  CH2 TRP  42      32.550  37.111  24.147
1.00 12.03
ATOM    390  C   TRP  42      32.851  37.625  31.433
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ATOM    391  O   TRP  42      32.485  36.498  31.755
1.00  9.69
ATOM    392  N   THR  43      33.084  38.597  32.309
1.00 10.28
ATOM    394  CA  THR  43      32.924  38.423  33.744
1.00 11.31
ATOM    395  CB  THR  43      33.385  39.688  34.487
1.00 13.67
ATOM    396  OG1 THR  43      34.716  40.025  34.065
1.00 16.25
ATOM    398  CG2 THR  43      33.393  39.450  35.988
1.00 15.27
ATOM    399  C   THR  43      31.477  38.108  34.115
1.00 11.22
ATOM    400  O   THR  43      31.219  37.325  35.032
1.00 11.38
ATOM    401  N   THR  44      30.541  38.720  33.407
1.00 11.08
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ATOM	403	CA	THR	44	29.132	38.479	33.658	
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ATOM	404	CB	THR	44	28.429	39.746	34.170	
1.00	13.88							
ATOM	405	OG1	THR	44	29.088	40.195	35.361	
1.00	16.33							
ATOM	407	CG2	THR	44	26.973	39.451	34.497	
1.00	15.15							
ATOM	408	C	THR	44	28.474	37.989	32.377	
1.00	10.65							
ATOM	409	O	THR	44	28.435	38.692	31.364	
1.00	11.08							
ATOM	410	N	GLY	45	28.034	36.742	32.416	
1.00	8.94							
ATOM	412	CA	GLY	45	27.381	36.161	31.267	
1.00	9.08							
ATOM	413	C	GLY	45	26.024	36.785	31.013	
1.00	8.92							
ATOM	414	O	GLY	45	25.426	37.413	31.888	
1.00	10.17							
ATOM	415	N	SER	46	25.528	36.587	29.803	
1.00	9.11							
ATOM	417	CA	SER	46	24.233	37.110	29.405	
1.00	9.90							
ATOM	418	CB	SER	46	24.421	38.433	28.649	
1.00	10.33							
ATOM	419	OG	SER	46	23.221	38.819	27.993	
1.00	11.62							
ATOM	421	C	SER	46	23.556	36.106	28.484	
1.00	9.38							
ATOM	422	O	SER	46	24.219	35.472	27.660	
1.00	9.60							
ATOM	423	N	PRO	47	22.232	35.919	28.641	
1.00	9.69							
ATOM	424	CD	PRO	47	21.363	36.467	29.703	
1.00	9.98							
ATOM	425	CA	PRO	47	21.500	34.985	27.788	
1.00	9.60							
ATOM	426	CB	PRO	47	20.190	34.774	28.559	
1.00	10.20							
ATOM	427	CG	PRO	47	19.978	36.084	29.224	
1.00	10.28							
ATOM	428	C	PRO	47	21.264	35.599	26.395	
1.00	9.80							
ATOM	429	O	PRO	47	20.771	34.915	25.492	
1.00	10.61							
ATOM	430	N	PHE	48	21.644	36.872	26.223	
1.00	8.32							
ATOM	432	CA	PHE	48	21.461	37.576	24.949	
1.00	8.97							
ATOM	433	CB	PHE	48	20.830	38.952	25.189	
1.00	10.54							
ATOM	434	CG	PHE	48	19.587	38.903	26.020	
1.00	12.40							
ATOM	435	CD1	PHE	48	18.507	38.142	25.610	
1.00	14.30							

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ATOM    436  CD2 PHE  48      19.527  39.558  27.239
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ATOM    437  CE1 PHE  48      17.379  38.028  26.410
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ATOM    438  CE2 PHE  48      18.400  39.448  28.043
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ATOM    439  CZ  PHE  48      17.332  38.683  27.627
1.00 14.77
ATOM    440  C   PHE  48      22.751  37.772  24.154
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ATOM    441  O   PHE  48      22.724  38.313  23.054
1.00  9.35
ATOM    442  N   ARG  49      23.876  37.324  24.699
1.00  8.82
ATOM    444  CA  ARG  49      25.152  37.512  24.019
1.00  8.97
ATOM    445  CB  ARG  49      26.306  37.267  24.992
1.00  9.78
ATOM    446  CG  ARG  49      27.663  37.691  24.421
1.00 10.95
ATOM    447  CD  ARG  49      28.794  37.437  25.386
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ATOM    448  NE  ARG  49      28.529  37.971  26.716
1.00 17.17
ATOM    450  CZ  ARG  49      28.663  39.245  27.065
1.00 16.53
ATOM    451  NH1 ARG  49      29.062  40.153  26.188
1.00 19.05
ATOM    454  NH2 ARG  49      28.418  39.606  28.314
1.00 18.90
ATOM    457  C   ARG  49      25.349  36.630  22.800
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ATOM    458  O   ARG  49      24.889  35.491  22.773
1.00  8.16
ATOM    459  N   THR  50      25.986  37.184  21.772
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ATOM    461  CA  THR  50      26.325  36.426  20.574
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ATOM    462  CB  THR  50      25.873  37.143  19.293
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ATOM    463  OG1 THR  50      24.453  37.318  19.334
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ATOM    465  CG2 THR  50      26.236  36.330  18.071
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ATOM    466  C   THR  50      27.849  36.345  20.636
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ATOM    467  O   THR  50      28.535  37.373  20.662
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ATOM    468  N   ILE  51      28.358  35.128  20.762
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ATOM    470  CA  ILE  51      29.789  34.895  20.881
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ATOM    471  CB  ILE  51      30.074  33.748  21.897
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ATOM    472  CG2 ILE  51      31.567  33.462  21.984
1.00 11.72
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ATOM	473	CG1	ILE	51	29.548	34.118	23.297	1.00 11.54
ATOM	474	CD1	ILE	51	28.097	33.689	23.565	1.00 11.67
ATOM	475	C	ILE	51	30.422	34.577	19.535	1.00 9.84
ATOM	476	O	ILE	51	29.891	33.784	18.753	1.00 9.89
ATOM	477	N	ASN	52	31.541	35.231	19.252	1.00 9.72
ATOM	479	CA	ASN	52	32.265	35.007	18.002	1.00 9.63
ATOM	480	CB	ASN	52	32.512	36.323	17.268	1.00 12.27
ATOM	481	CG	ASN	52	31.239	37.060	16.956	1.00 16.92
ATOM	482	OD1	ASN	52	30.579	36.770	15.960	1.00 20.42
ATOM	483	ND2	ASN	52	30.869	38.004	17.817	1.00 18.67
ATOM	486	C	ASN	52	33.618	34.430	18.359	1.00 8.99
ATOM	487	O	ASN	52	34.216	34.813	19.369	1.00 8.30
ATOM	488	N	TYR	53	34.101	33.502	17.546	1.00 8.54
ATOM	490	CA	TYR	53	35.417	32.938	17.805	1.00 8.38
ATOM	491	CB	TYR	53	35.383	31.888	18.931	1.00 8.90
ATOM	492	CG	TYR	53	34.816	30.545	18.510	1.00 8.55
ATOM	493	CD1	TYR	53	35.652	29.541	18.013	1.00 8.13
ATOM	494	CE1	TYR	53	35.142	28.318	17.595	1.00 7.11
ATOM	495	CD2	TYR	53	33.446	30.286	18.582	1.00 8.30
ATOM	496	CE2	TYR	53	32.924	29.062	18.164	1.00 7.39
ATOM	497	CZ	TYR	53	33.771	28.085	17.670	1.00 7.17
ATOM	498	OH	TYR	53	33.270	26.875	17.235	1.00 8.09
ATOM	500	C	TYR	53	35.995	32.333	16.549	1.00 8.13
ATOM	501	O	TYR	53	35.289	32.095	15.567	1.00 8.60
ATOM	502	N	ASN	54	37.305	32.146	16.568	1.00 7.89
ATOM	504	CA	ASN	54	38.011	31.525	15.466	1.00 8.58
ATOM	505	CB	ASN	54	38.628	32.583	14.532	1.00 8.69
ATOM	506	CG	ASN	54	39.273	31.967	13.291	1.00 9.70

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ATOM    507  OD1 ASN  54      39.001  32.391  12.157
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ATOM    508  ND2 ASN  54      40.133  30.989  13.490
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ATOM    511  C   ASN  54      39.106  30.705  16.137
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ATOM    512  O   ASN  54      39.984  31.262  16.807
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ATOM    513  N   ALA  55      39.010  29.386  16.031
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ATOM    515  CA  ALA  55      40.028  28.510  16.607
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ATOM    516  CB  ALA  55      39.406  27.201  17.109
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ATOM    517  C   ALA  55      41.090  28.235  15.529
1.00  9.13
ATOM    518  O   ALA  55      40.897  27.380  14.656
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ATOM    519  N   GLY  56      42.176  29.000  15.569
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ATOM    521  CA  GLY  56      43.254  28.829  14.606
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ATOM    522  C   GLY  56      43.912  27.459  14.705
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ATOM    523  O   GLY  56      44.430  26.928  13.716
1.00 11.91
ATOM    524  N   VAL  57      43.933  26.900  15.910
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ATOM    526  CA  VAL  57      44.483  25.572  16.150
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ATOM    527  CB  VAL  57      45.869  25.628  16.856
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ATOM    528  CG1 VAL  57      46.378  24.211  17.158
1.00 10.95
ATOM    529  CG2 VAL  57      46.873  26.378  15.998
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ATOM    530  C   VAL  57      43.530  24.837  17.086
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ATOM    531  O   VAL  57      43.121  25.388  18.118
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ATOM    532  N   TRP  58      43.104  23.644  16.682
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ATOM    534  CA  TRP  58      42.258  22.797  17.520
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ATOM    535  CB  TRP  58      40.771  22.855  17.137
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ATOM    536  CG  TRP  58      39.950  21.927  17.984
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ATOM    537  CD2 TRP  58      39.906  21.883  19.421
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ATOM    538  CE2 TRP  58      39.101  20.778  19.781
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ATOM    539  CE3 TRP  58      40.478  22.662  20.434
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ATOM    540  CD1 TRP  58      39.177  20.883  17.550
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ATOM    544  CZ3 TRP    58      40.234  22.321  21.756
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ATOM    545  CH2 TRP    58      39.428  21.214  22.083
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ATOM    546  C   TRP    58      42.841  21.418  17.283
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ATOM    547  O   TRP    58      42.496  20.741  16.323
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ATOM    551  CB  ALA    59      45.850  20.038  17.520
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ATOM    552  C   ALA    59      44.503  18.893  19.295
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ATOM    553  O   ALA    59      45.574  18.702  19.893
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ATOM    554  N   PRO    60      43.345  18.417  19.763
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ATOM    555  CD  PRO    60      41.964  18.696  19.313
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ATOM    556  CA  PRO    60      43.355  17.582  20.963
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ATOM    557  CB  PRO    60      41.878  17.587  21.386
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ATOM    558  CG  PRO    60      41.157  17.657  20.069
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ATOM    559  C   PRO    60      43.827  16.165  20.610
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ATOM    560  O   PRO    60      43.472  15.638  19.559
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ATOM    561  N   ASN    61      44.655  15.580  21.470
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ATOM    564  CB  ASN    61      46.663  14.142  21.519
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ATOM    565  CG  ASN    61      47.422  14.081  20.239
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ATOM    566  OD1 ASN    61      46.846  14.130  19.159
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ATOM    567  ND2 ASN    61      48.733  13.960  20.342
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ATOM    570  C   ASN    61      44.502  13.396  22.418
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ATOM    571  O   ASN    61      45.193  12.805  23.265
1.00 20.86
ATOM    572  N   GLY    62      43.181  13.442  22.478
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ATOM    574  CA  GLY    62      42.460  12.724  23.505
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ATOM    575  C   GLY  62      41.195  13.489  23.824
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ATOM    576  O   GLY  62      40.695  14.229  22.976
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ATOM    577  N   ASN  63      40.733  13.379  25.064
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ATOM    579  CA  ASN  63      39.501  14.033  25.508
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ATOM    580  CB  ASN  63      38.994  13.326  26.769
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ATOM    581  CG  ASN  63      37.598  13.776  27.201
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ATOM    582  OD1 ASN  63      37.040  13.221  28.147
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ATOM    583  ND2 ASN  63      37.033  14.768  26.529
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ATOM    586  C   ASN  63      39.673  15.531  25.764
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ATOM    588  N   GLY  64      39.040  16.339  24.919
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ATOM    590  CA  GLY  64      39.092  17.787  25.045
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ATOM    591  C   GLY  64      37.887  18.376  24.327
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ATOM    592  O   GLY  64      37.430  17.794  23.339
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ATOM    593  N   TYR  65      37.366  19.496  24.830
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ATOM    595  CA  TYR  65      36.198  20.172  24.242
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ATOM    596  CB  TYR  65      35.032  20.281  25.249
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ATOM    597  CG  TYR  65      34.628  19.022  25.992
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ATOM    598  CD1 TYR  65      34.618  17.780  25.369
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ATOM    599  CE1 TYR  65      34.220  16.645  26.048
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ATOM    600  CD2 TYR  65      34.222  19.090  27.324
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ATOM    601  CE2 TYR  65      33.816  17.959  28.012
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ATOM    602  CZ  TYR  65      33.817  16.739  27.370
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ATOM    603  OH  TYR  65      33.412  15.599  28.045
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ATOM    605  C   TYR  65      36.520  21.619  23.866
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ATOM    606  O   TYR  65      37.244  22.316  24.588
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ATOM    607  N   LEU  66      35.970  22.063  22.743
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ATOM    609  CA  LEU  66      36.083  23.445  22.283
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ATOM    611  CG  LEU  66      36.461  24.907  20.208
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ATOM    612  CD1 LEU  66      37.475  25.780  20.976
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ATOM    613  CD2 LEU  66      36.803  24.829  18.727
1.00  9.90
ATOM    614  C   LEU  66      34.622  23.830  22.525
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ATOM    615  O   LEU  66      33.722  23.435  21.758
1.00  5.88
ATOM    616  N   THR  67      34.389  24.653  23.542
1.00  6.32
ATOM    618  CA  THR  67      33.017  24.899  23.964
1.00  6.62
ATOM    619  CB  THR  67      32.687  23.771  25.014
1.00  5.96
ATOM    620  OG1 THR  67      31.338  23.878  25.482
1.00  6.25
ATOM    622  CG2 THR  67      33.641  23.834  26.239
1.00  6.12
ATOM    623  C   THR  67      32.757  26.231  24.650
1.00  5.91
ATOM    624  O   THR  67      33.651  26.792  25.281
1.00  5.87
ATOM    625  N   LEU  68      31.541  26.756  24.496
1.00  5.49
ATOM    627  CA  LEU  68      31.141  27.950  25.232
1.00  5.70
ATOM    628  CB  LEU  68      29.806  28.488  24.741
1.00  7.05
ATOM    629  CG  LEU  68      29.314  29.725  25.491
1.00  8.06
ATOM    630  CD1 LEU  68      30.210  30.922  25.209
1.00 10.31
ATOM    631  CD2 LEU  68      27.874  30.009  25.092
1.00 10.13
ATOM    632  C   LEU  68      30.961  27.334  26.626
1.00  6.24
ATOM    633  O   LEU  68      30.436  26.212  26.765
1.00  6.15
ATOM    634  N   TYR  69      31.357  28.062  27.656
1.00  5.92
ATOM    636  CA  TYR  69      31.323  27.516  28.997
1.00  6.43
ATOM    637  CB  TYR  69      32.733  26.961  29.292
1.00  7.92
ATOM    638  CG  TYR  69      32.942  26.431  30.685
1.00  7.81
ATOM    639  CD1 TYR  69      32.482  25.170  31.039
1.00  8.51
ATOM    640  CE1 TYR  69      32.650  24.689  32.315
1.00  9.90
ATOM    641  CD2 TYR  69      33.586  27.199  31.652
1.00  8.54
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ATOM    642  CE2 TYR  69      33.756  26.727  32.931
1.00  8.83
ATOM    643  CZ  TYR  69      33.282  25.472  33.258
1.00  8.64
ATOM    644  OH  TYR  69      33.410  25.015  34.551
1.00 10.08
ATOM    646  C   TYR  69      30.964  28.601  29.989
1.00  6.66
ATOM    647  O   TYR  69      31.456  29.723  29.898
1.00  6.52
ATOM    648  N   GLY  70      30.119  28.274  30.957
1.00  5.93
ATOM    650  CA  GLY  70      29.770  29.283  31.935
1.00  7.20
ATOM    651  C   GLY  70      29.001  28.724  33.108
1.00  6.61
ATOM    652  O   GLY  70      28.698  27.530  33.167
1.00  6.24
ATOM    653  N   TRP  71      28.699  29.602  34.054
1.00  7.21
ATOM    655  CA  TRP  71      27.953  29.215  35.240
1.00  7.25
ATOM    656  CB  TRP  71      28.865  29.170  36.478
1.00  8.08
ATOM    657  CG  TRP  71      29.887  28.097  36.524
1.00  9.15
ATOM    658  CD2 TRP  71      29.803  26.869  37.253
1.00  9.01
ATOM    659  CE2 TRP  71      31.016  26.176  37.049
1.00  9.87
ATOM    660  CE3 TRP  71      28.818  26.283  38.057
1.00 10.02
ATOM    661  CD1 TRP  71      31.112  28.106  35.920
1.00  8.19
ATOM    662  NE1 TRP  71      31.793  26.952  36.228
1.00  9.71
ATOM    664  CZ2 TRP  71      31.271  24.925  37.621
1.00 10.26
ATOM    665  CZ3 TRP  71      29.068  25.043  38.621
1.00 11.63
ATOM    666  CH2 TRP  71      30.286  24.375  38.401
1.00 11.11
ATOM    667  C   TRP  71      26.893  30.220  35.620
1.00  7.86
ATOM    668  O   TRP  71      26.976  31.401  35.273
1.00  7.90
ATOM    669  N   THR  72      25.895  29.725  36.340
1.00  9.00
ATOM    671  CA  THR  72      24.907  30.593  36.960
1.00  9.70
ATOM    672  CB  THR  72      23.473  30.521  36.379
1.00 10.06
ATOM    673  OG1 THR  72      22.939  29.204  36.544
1.00 10.75
ATOM    675  CG2 THR  72      23.431  30.955  34.929
1.00  9.49
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ATOM    677  O   THR  72      25.246  28.926  38.670
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ATOM    678  N   ARG  73      24.362  30.914  39.289
1.00 13.21
ATOM    680  CA  ARG  73      24.156  30.555  40.687
1.00 15.05
ATOM    681  CB  ARG  73      24.932  31.488  41.621
1.00 16.77
ATOM    682  CG  ARG  73      26.430  31.290  41.596
1.00 18.20
ATOM    683  CD  ARG  73      27.103  32.107  42.682
1.00 19.22
ATOM    684  NE  ARG  73      28.538  31.835  42.766
1.00 21.45
ATOM    686  CZ  ARG  73      29.466  32.495  42.079
1.00 22.40
ATOM    687  NH1 ARG  73      29.118  33.465  41.247
1.00 24.34
ATOM    690  NH2 ARG  73      30.748  32.209  42.247
1.00 23.90
ATOM    693  C   ARG  73      22.658  30.745  40.901
1.00 15.76
ATOM    694  O   ARG  73      22.001  31.435  40.114
1.00 15.54
ATOM    695  N   SER  74      22.125  30.124  41.949
1.00 17.09
ATOM    697  CA  SER  74      20.701  30.215  42.283
1.00 18.69
ATOM    698  CB  SER  74      20.370  31.620  42.798
1.00 19.69
ATOM    699  OG  SER  74      21.282  31.998  43.817
1.00 24.14
ATOM    701  C   SER  74      19.759  29.838  41.126
1.00 17.91
ATOM    702  O   SER  74      18.998  30.673  40.625
1.00 18.35
ATOM    703  N   PRO  75      19.793  28.572  40.693
1.00 17.77
ATOM    704  CD  PRO  75      18.753  28.012  39.810
1.00 18.55
ATOM    705  CA  PRO  75      20.653  27.508  41.224
1.00 17.24
ATOM    706  CB  PRO  75      19.828  26.253  40.969
1.00 18.06
ATOM    707  CG  PRO  75      19.175  26.565  39.666
1.00 18.68
ATOM    708  C   PRO  75      21.999  27.423  40.515
1.00 16.14
ATOM    709  O   PRO  75      22.231  28.081  39.492
1.00 15.03
ATOM    710  N   LEU  76      22.875  26.608  41.080
1.00 14.58
ATOM    712  CA  LEU  76      24.207  26.389  40.547
1.00 14.02
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ATOM    713  CB  LEU  76      25.063  25.722  41.623
1.00 16.02
ATOM    714  CG  LEU  76      26.575  25.774  41.487
1.00 18.13
ATOM    715  CD1 LEU  76      27.047  27.219  41.526
1.00 18.22
ATOM    716  CD2 LEU  76      27.197  24.970  42.626
1.00 18.98
ATOM    717  C   LEU  76      24.112  25.488  39.312
1.00 12.79
ATOM    718  O   LEU  76      23.722  24.323  39.412
1.00 12.48
ATOM    719  N   ILE  77      24.482  26.025  38.151
1.00 11.37
ATOM    721  CA  ILE  77      24.431  25.266  36.902
1.00 10.64
ATOM    722  CB  ILE  77      23.188  25.655  36.054
1.00 11.91
ATOM    723  CG2 ILE  77      23.253  24.994  34.686
1.00 11.77
ATOM    724  CG1 ILE  77      21.898  25.234  36.769
1.00 12.91
ATOM    725  CD1 ILE  77      20.656  25.787  36.160
1.00 14.88
ATOM    726  C   ILE  77      25.676  25.555  36.068
1.00  8.64
ATOM    727  O   ILE  77      26.084  26.712  35.948
1.00  8.92
ATOM    728  N   GLU  78      26.319  24.502  35.574
1.00  7.66
ATOM    730  CA  GLU  78      27.503  24.621  34.720
1.00  7.21
ATOM    731  CB  GLU  78      28.536  23.573  35.134
1.00  7.73
ATOM    732  CG  GLU  78      29.868  23.635  34.372
1.00  9.08
ATOM    733  CD  GLU  78      30.835  22.544  34.805
1.00 10.20
ATOM    734  OE1 GLU  78      32.050  22.814  34.932
1.00 10.78
ATOM    735  OE2 GLU  78      30.395  21.407  35.026
1.00 10.70
ATOM    736  C   GLU  78      26.961  24.315  33.322
1.00  7.93
ATOM    737  O   GLU  78      26.391  23.240  33.112
1.00  9.13
ATOM    738  N   TYR  79      27.119  25.229  32.369
1.00  7.30
ATOM    740  CA  TYR  79      26.562  24.982  31.046
1.00  6.18
ATOM    741  CB  TYR  79      25.422  25.963  30.737
1.00  6.50
ATOM    742  CG  TYR  79      25.844  27.414  30.631
1.00  6.96
ATOM    743  CD1 TYR  79      26.466  27.896  29.479
1.00  6.30
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ATOM    744  CE1 TYR  79      26.861  29.215  29.376
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ATOM    745  CD2 TYR  79      25.626  28.299  31.681
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ATOM    746  CE2 TYR  79      26.019  29.633  31.584
1.00  7.44
ATOM    747  CZ  TYR  79      26.636  30.079  30.427
1.00  7.25
ATOM    748  OH  TYR  79      27.042  31.388  30.303
1.00  8.96
ATOM    750  C   TYR  79      27.605  24.969  29.939
1.00  7.01
ATOM    751  O   TYR  79      28.658  25.619  30.044
1.00  5.99
ATOM    752  N   TYR  80      27.235  24.315  28.840
1.00  6.31
ATOM    754  CA  TYR  80      28.112  24.128  27.692
1.00  5.86
ATOM    755  CB  TYR  80      28.667  22.695  27.722
1.00  5.72
ATOM    756  CG  TYR  80      29.620  22.352  28.827
1.00  5.27
ATOM    757  CD1 TYR  80      29.159  21.881  30.055
1.00  7.53
ATOM    758  CE1 TYR  80      30.042  21.465  31.032
1.00  7.89
ATOM    759  CD2 TYR  80      30.983  22.403  28.608
1.00  7.64
ATOM    760  CE2 TYR  80      31.865  21.995  29.566
1.00  8.52
ATOM    761  CZ  TYR  80      31.394  21.523  30.776
1.00  9.81
ATOM    762  OH  TYR  80      32.297  21.100  31.725
1.00 10.19
ATOM    764  C   TYR  80      27.413  24.207  26.346
1.00  6.25
ATOM    765  O   TYR  80      26.268  23.762  26.216
1.00  6.36
ATOM    766  N   VAL  81      28.111  24.755  25.354
1.00  5.60
ATOM    768  CA  VAL  81      27.650  24.709  23.956
1.00  4.96
ATOM    769  CB  VAL  81      27.189  26.044  23.369
1.00  6.50
ATOM    770  CG1 VAL  81      26.812  25.814  21.903
1.00  6.96
ATOM    771  CG2 VAL  81      25.955  26.567  24.133
1.00  7.32
ATOM    772  C   VAL  81      28.915  24.204  23.252
1.00  5.73
ATOM    773  O   VAL  81      29.854  24.963  22.970
1.00  5.99
ATOM    774  N   VAL  82      28.957  22.897  23.048
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ATOM    776  CA  VAL  82      30.123  22.242  22.470
1.00  5.85
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ATOM 777 CB VAL 82 30.246 20.785 23.010
1.00 5.93
ATOM 778 CG1 VAL 82 31.540 20.140 22.517
1.00 7.63
ATOM 779 CG2 VAL 82 30.205 20.766 24.539
1.00 6.83
ATOM 780 C VAL 82 30.119 22.221 20.951
1.00 6.61
ATOM 781 O VAL 82 29.261 21.582 20.348
1.00 6.74
ATOM 782 N ASP 83 31.119 22.864 20.340
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ATOM 784 CA ASP 83 31.237 22.909 18.874
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ATOM 785 CB ASP 83 31.679 24.290 18.395
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ATOM 786 CG ASP 83 30.504 25.196 18.052
1.00 6.35
ATOM 787 OD1 ASP 83 30.737 26.382 17.772
1.00 7.42
ATOM 788 OD2 ASP 83 29.350 24.720 18.044
1.00 7.63
ATOM 789 C ASP 83 32.160 21.835 18.312
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ATOM 790 O ASP 83 31.995 21.399 17.175
1.00 7.51
ATOM 791 N SER 84 33.188 21.479 19.069
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ATOM 793 CA SER 84 34.085 20.424 18.641
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ATOM 794 CB SER 84 35.198 20.933 17.727
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ATOM 795 OG SER 84 35.909 19.816 17.225
1.00 6.88
ATOM 797 C SER 84 34.655 19.706 19.851
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ATOM 798 O SER 84 34.527 20.167 20.988
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ATOM 799 N TRP 85 35.236 18.545 19.606
1.00 6.77
ATOM 801 CA TRP 85 35.780 17.732 20.675
1.00 7.16
ATOM 802 CB TRP 85 34.667 16.856 21.276
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ATOM 803 CG TRP 85 33.955 15.981 20.279
1.00 8.40
ATOM 804 CD2 TRP 85 32.790 16.319 19.504
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ATOM 805 CE2 TRP 85 32.500 15.209 18.683
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ATOM 806 CE3 TRP 85 31.970 17.449 19.425
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ATOM 807 CD1 TRP 85 34.303 14.711 19.911
1.00 9.75
ATOM 808 NE1 TRP 85 33.434 14.243 18.950
1.00 10.01

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ATOM    810  CZ2 TRP    85      31.428  15.198  17.793
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ATOM    811  CZ3 TRP    85      30.905  17.432  18.537
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ATOM    812  CH2 TRP    85      30.649  16.313  17.737
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ATOM    813  C   TRP    85      36.914  16.881  20.127
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ATOM    814  O   TRP    85      37.254  16.976  18.939
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ATOM    815  N   GLY    86      37.518  16.094  21.006
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ATOM    817  CA  GLY    86      38.603  15.225  20.609
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ATOM    818  C   GLY    86      38.137  13.815  20.335
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ATOM    819  O   GLY    86      37.402  13.561  19.381
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ATOM    820  N   THR    87      38.534  12.895  21.197
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ATOM    822  CA  THR    87      38.185  11.491  21.029
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ATOM    823  CB  THR    87      39.277  10.586  21.643
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ATOM    824  OG1 THR    87      39.518  10.996  22.996
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ATOM    826  CG2 THR    87      40.579  10.676  20.842
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ATOM    827  C   THR    87      36.852  11.086  21.637
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ATOM    828  O   THR    87      36.345   9.997  21.350
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ATOM    829  N   TYR    88      36.286  11.949  22.473
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ATOM    831  CA  TYR    88      35.033  11.641  23.165
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ATOM    832  CB  TYR    88      35.323  11.459  24.669
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ATOM    833  CG  TYR    88      34.096  11.360  25.553
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ATOM    834  CD1 TYR    88      33.282  10.236  25.521
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ATOM    835  CE1 TYR    88      32.116  10.177  26.273
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ATOM    836  CD2 TYR    88      33.720  12.419  26.372
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ATOM    837  CE2 TYR    88      32.566  12.368  27.127
1.00 17.85
ATOM    838  CZ  TYR    88      31.763  11.250  27.071
1.00 19.24
ATOM    839  OH  TYR    88      30.583  11.222  27.789
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ATOM    841  C   TYR    88      33.970  12.713  22.981
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ATOM    842  O   TYR    88      34.211  13.886  23.290
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ATOM    845  CA  ARG  89      31.717  13.277  22.278
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ATOM    846  CB  ARG  89      30.852  12.918  21.063
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ATOM    847  CG  ARG  89      29.751  13.920  20.805
1.00  8.70
ATOM    848  CD  ARG  89      28.867  13.540  19.623
1.00  8.30
ATOM    849  NE  ARG  89      27.824  14.550  19.439
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ATOM    851  CZ  ARG  89      26.565  14.414  19.857
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ATOM    852  NH1 ARG  89      26.170  13.300  20.460
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ATOM    855  NH2 ARG  89      25.729  15.435  19.750
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ATOM    858  C   ARG  89      30.886  13.218  23.563
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ATOM    859  O   ARG  89      30.381  12.157  23.936
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ATOM    860  N   PRO  90      30.766  14.348  24.276
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ATOM    861  CD  PRO  90      31.298  15.695  23.987
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ATOM    862  CA  PRO  90      29.984  14.337  25.514
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ATOM    863  CB  PRO  90      30.311  15.695  26.139
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ATOM    864  CG  PRO  90      30.543  16.570  24.965
1.00 10.45
ATOM    865  C   PRO  90      28.486  14.136  25.294
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ATOM    866  O   PRO  90      27.890  14.725  24.385
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ATOM    867  N   THR  91      27.890  13.263  26.096
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ATOM    869  CA  THR  91      26.459  13.001  25.995
1.00 13.17
ATOM    870  CB  THR  91      26.144  11.723  25.167
1.00 13.69
ATOM    871  OG1 THR  91      26.655  10.563  25.841
1.00 16.21
ATOM    873  CG2 THR  91      26.757  11.814  23.754
1.00 13.50
ATOM    874  C   THR  91      25.885  12.875  27.406
1.00 13.53
ATOM    875  O   THR  91      26.626  12.903  28.392
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ATOM    876  N   GLY  92      24.566  12.783  27.505
1.00 12.71
ATOM    878  CA  GLY  92      23.915  12.659  28.798
1.00 12.21
ATOM    879  C   GLY  92      22.488  12.240  28.541
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ATOM    880  O   GLY   92      22.232  11.459  27.617
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ATOM    881  N   THR   93      21.558  12.754  29.343
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ATOM    883  CA  THR   93      20.143  12.442  29.175
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ATOM    884  CB  THR   93      19.380  12.724  30.480
1.00 13.13
ATOM    885  OG1 THR   93      19.941  11.922  31.528
1.00 14.32
ATOM    887  CG2 THR   93      17.897  12.397  30.328
1.00 14.22
ATOM    888  C   THR   93      19.606  13.311  28.039
1.00 10.68
ATOM    889  O   THR   93      19.596  14.538  28.139
1.00 10.38
ATOM    890  N   TYR   94      19.191  12.668  26.951
1.00 10.81
ATOM    892  CA  TYR   94      18.691  13.369  25.775
1.00 10.59
ATOM    893  CB  TYR   94      18.532  12.381  24.619
1.00 11.92
ATOM    894  CG  TYR   94      18.014  13.008  23.347
1.00 11.53
ATOM    895  CD1 TYR   94  .00  8.59
ATOM    964  CG  ASP  101      24.966  30.278  15.615
1.00  9.23
ATOM    965  OD1 ASP  101      24.632  29.849  16.740
1.00  8.01
ATOM    966  OD2 ASP  101      24.891  31.481  15.300
1.00 11.33
ATOM    967  C   ASP  101      24.872  27.940  12.547
1.00 10.53
ATOM    968  O   ASP  101      26.063  27.625  12.533
1.00 11.20
ATOM    969  N   GLY  102      23.957  27.394  11.746
1.00 10.95
ATOM    971  CA  GLY  102      24.323  26.415  10.740
1.00 11.74
ATOM    972  C   GLY  102      24.522  24.991  11.203
1.00 12.03
ATOM    973  O   GLY  102      24.849  24.130  10.387
1.00 15.13
ATOM    974  N   GLY  103      24.331  24.720  12.489
1.00 10.73
ATOM    976  CA  GLY  103      24.520  23.366  12.968
1.00  9.93
ATOM    977  C   GLY  103      23.331  22.865  13.753
1.00  8.93
ATOM    978  O   GLY  103      22.442  23.643  14.096
1.00  9.85
ATOM    979  N   THR  104      23.272  21.554  13.952
1.00  8.69
ATOM    981  CA  THR  104      22.207  20.950  14.753
1.00  8.72
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ATOM    982  CB  THR  104      21.513  19.802  14.014
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ATOM    983  OG1 THR  104      20.972  20.313  12.788
1.00 13.06
ATOM    985  CG2 THR  104      20.379  19.211  14.873
1.00 10.48
ATOM    986  C   THR  104      22.887  20.429  16.020
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ATOM    987  O   THR  104      23.919  19.741  15.948
1.00  8.43
ATOM    988  N   TYR  105      22.312  20.766  17.167
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ATOM    990  CA  TYR  105      22.869  20.382  18.453
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ATOM    991  CB  TYR  105      23.138  21.636  19.296
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ATOM    992  CG  TYR  105      24.168  22.602  18.740
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ATOM    993  CD1 TYR  105      23.853  23.459  17.693
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ATOM    994  CE1 TYR  105      24.777  24.367  17.210
1.00  7.90
ATOM    995  CD2 TYR  105      25.447  22.677  19.292
1.00  6.18
ATOM    996  CE2 TYR  105      26.379  23.585  18.818
1.00  7.32
ATOM    997  CZ  TYR  105      26.036  24.425  17.779
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ATOM    998  OH  TYR  105      26.940  25.358  17.306
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ATOM   1000  C   TYR  105      21.907  19.505  19.240
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ATOM   1001  O   TYR  105      20.703  19.742  19.233
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ATOM   1002  N   ASP  106      22.436  18.470  19.877
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ATOM   1004  CA  ASP  106      21.621  17.629  20.744
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ATOM   1005  CB  ASP  106      22.232  16.242  20.895
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ATOM   1006  CG  ASP  106      22.135  15.434  19.635
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ATOM   1007  OD1 ASP  106      23.134  14.788  19.269
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ATOM   1008  OD2 ASP  106      21.051  15.468  19.011
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ATOM   1009  C   ASP  106      21.650  18.319  22.100
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ATOM   1010  O   ASP  106      22.678  18.897  22.489
1.00  8.85
ATOM   1011  N   ILE  107      20.545  18.254  22.830
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ATOM   1014  CB  ILE  107      19.151  19.675  24.297
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ATOM   1016  CG1 ILE   107      19.117  20.790  23.249
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ATOM   1017  CD1 ILE   107      17.748  21.473  23.124
1.00  9.19
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ATOM   1019  O   ILE   107      19.722  16.789  25.123
1.00  7.22
ATOM   1020  N   TYR   108      21.389  17.880  26.173
1.00  6.95
ATOM   1022  CA  TYR   108      21.515  16.882  27.232
1.00  7.18
ATOM   1023  CB  TYR   108      22.811  16.077  27.090
1.00  7.59
ATOM   1024  CG  TYR   108      23.013  15.341  25.794
1.00  8.26
ATOM   1025  CD1 TYR   108      23.988  15.753  24.887
1.00  8.13
ATOM   1026  CE1 TYR   108      24.213  15.060  23.719
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ATOM   1027  CD2 TYR   108      22.267  14.208  25.493
1.00  8.68
ATOM   1028  CE2 TYR   108      22.486  13.502  24.324
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ATOM   1029  CZ  TYR   108      23.458  13.933  23.441
1.00  9.66
ATOM   1030  OH  TYR   108      23.681  13.242  22.276
1.00  9.55
ATOM   1032  C   TYR   108      21.617  17.506  28.607
1.00  7.18
ATOM   1033  O   TYR   108      22.041  18.662  28.748
1.00  6.50
ATOM   1034  N   THR   109      21.259  16.721  29.620
1.00  7.71
ATOM   1036  CA  THR   109      21.465  17.144  31.003
1.00  8.74
ATOM   1037  CB  THR   109      20.173  17.372  31.831
1.00  9.86
ATOM   1038  OG1 THR   109      19.393  16.169  31.862
1.00 11.56
ATOM   1040  CG2 THR   109      19.371  18.521  31.276
1.00 12.46
ATOM   1041  C   THR   109      22.241  16.028  31.670
1.00  8.80
ATOM   1042  O   THR   109      22.121  14.849  31.295
1.00  8.91
ATOM   1043  N   THR   110      23.095  16.412  32.606
1.00  8.78
ATOM   1045  CA  THR   110      23.876  15.470  33.401
1.00 10.13
ATOM   1046  CB  THR   110      25.327  15.244  32.854
1.00 10.63
ATOM   1047  OG1 THR   110      26.021  16.493  32.771
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ATOM   1050  C   THR   110      23.972  16.094  34.792
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ATOM   1051  O   THR   110      23.632  17.264  34.990
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ATOM   1052  N   THR   111      24.451  15.312  35.747
1.00 10.72
ATOM   1054  CA  THR   111      24.620  15.777  37.114
1.00 11.19
ATOM   1055  CB  THR   111      23.582  15.125  38.047
1.00 13.02
ATOM   1056  OG1 THR   111      22.270  15.388  37.539
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ATOM   1058  CG2 THR   111      23.695  15.684  39.476
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ATOM   1059  C   THR   111      26.015  15.372  37.566
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ATOM   1060  O   THR   111      26.505  14.306  37.199
1.00 11.90
ATOM   1061  N   ARG   112      26.674  16.269  38.290
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ATOM   1063  CA  ARG   112      28.002  16.016  38.839
1.00 11.90
ATOM   1064  CB  ARG   112      28.930  17.201  38.558
1.00 14.38
ATOM   1065  CG  ARG   112      29.291  17.330  37.103
1.00 16.14
ATOM   1066  CD  ARG   112      30.426  16.404  36.731
1.00 19.66
ATOM   1067  NE  ARG   112      31.707  17.057  36.981
1.00 23.60
ATOM   1069  CZ  ARG   112      32.767  16.477  37.542
1.00 25.66
ATOM   1070  NH1 ARG   112      32.724  15.204  37.927
1.00 27.05
ATOM   1073  NH2 ARG   112      33.867  17.193  37.756
1.00 27.17
ATOM   1076  C   ARG   112      27.819  15.843  40.344
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ATOM   1077  O   ARG   112      27.071  16.595  40.978
1.00 11.70
ATOM   1078  N   TYR   113      28.465  14.829  40.900
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ATOM   1080  CA  TYR   113      28.366  14.530  42.323
1.00 12.45
ATOM   1081  CB  TYR   113      27.883  13.090  42.519
1.00 12.53
ATOM   1082  CG  TYR   113      26.550  12.818  41.855
1.00 13.06
ATOM   1083  CD1 TYR   113      25.356  13.014  42.547
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ATOM   1084  CE1 TYR   113      24.139  12.792  41.945
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ATOM   1085  CD2 TYR   113      26.489  12.389  40.536
1.00 13.33
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ATOM   1086  CE2 TYR   113      25.267  12.162  39.920
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ATOM   1087  CZ  TYR   113      24.099  12.365  40.635
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ATOM   1088  OH  TYR   113      22.884  12.113  40.036
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ATOM   1090  C   TYR   113      29.702  14.724  43.032
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ATOM   1091  O   TYR   113      30.718  14.139  42.625
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ATOM   1092  N   ASN   114      29.678  15.537  44.088
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ATOM   1094  CA  ASN   114      30.861  15.840  44.901
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ATOM   1095  CB  ASN   114      31.251  14.610  45.723
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ATOM   1096  CG  ASN   114      32.206  14.942  46.855
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ATOM   1097  OD1 ASN   114      32.035  15.938  47.562
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ATOM   1098  ND2 ASN   114      33.191  14.085  47.057
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ATOM   1101  C   ASN   114      32.017  16.274  44.000
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ATOM   1102  O   ASN   114      33.127  15.736  44.073
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ATOM   1103  N   ALA   115      31.743  17.264  43.162
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ATOM   1105  CA  ALA   115      32.726  17.764  42.208
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ATOM   1106  CB  ALA   115      32.176  17.594  40.786
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ATOM   1107  C   ALA   115      33.037  19.231  42.471
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ATOM   1108  O   ALA   115      32.214  19.955  43.039
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ATOM   1109  N   PRO   116      34.232  19.693  42.055
1.00 12.08
ATOM   1110  CD  PRO   116      35.306  18.936  41.388
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ATOM   1111  CA  PRO   116      34.619  21.092  42.258
1.00 12.75
ATOM   1112  CB  PRO   116      36.022  21.158  41.642
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ATOM   1113  CG  PRO   116      36.525  19.757  41.718
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ATOM   1114  C   PRO   116      33.666  22.013  41.496
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ATOM   1115  O   PRO   116      33.319  21.745  40.339
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ATOM   1116  N   SER   117      33.228  23.084  42.142
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ATOM   1118  CA  SER   117      32.339  24.019  41.482
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ATOM   1119  CB  SER   117      31.012  24.124  42.231
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ATOM   1120  OG  SER 117      31.158  24.930  43.385
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ATOM   1122  C   SER 117      33.022  25.379  41.436
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ATOM   1123  O   SER 117      34.129  25.553  41.960
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ATOM   1124  N   ILE 118      32.328  26.348  40.853
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ATOM   1126  CA  ILE 118      32.837  27.703  40.728
1.00 16.31
ATOM   1127  CB  ILE 118      31.813  28.600  39.976
1.00 15.37
ATOM   1128  CG2 ILE 118      30.488  28.670  40.731
1.00 14.25
ATOM   1129  CG1 ILE 118      32.392  29.993  39.741
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ATOM   1130  CD1 ILE 118      31.520  30.875  38.868
1.00 16.65
ATOM   1131  C   ILE 118      33.206  28.321  42.080
1.00 17.92
ATOM   1132  O   ILE 118      34.183  29.055  42.174
1.00 18.87
ATOM   1133  N   ASP 119      32.455  27.974  43.123
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ATOM   1135  CA  ASP 119      32.674  28.507  44.472
1.00 22.48
ATOM   1136  CB  ASP 119      31.415  28.308  45.327
1.00 24.91
ATOM   1137  CG  ASP 119      30.283  29.248  44.952
1.00 26.95
ATOM   1138  OD1 ASP 119      30.568  30.400  44.566
1.00 27.78
ATOM   1139  OD2 ASP 119      29.106  28.830  45.071
1.00 29.45
ATOM   1140  C   ASP 119      33.834  27.866  45.226
1.00 23.22
ATOM   1141  O   ASP 119      34.165  28.300  46.339
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ATOM   1142  N   GLY 120      34.450  26.844  44.640
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ATOM   1144  CA  GLY 120      35.519  26.148  45.332
1.00 21.06
ATOM   1145  C   GLY 120      34.867  24.995  46.081
1.00 20.24
ATOM   1146  O   GLY 120      33.639  24.838  46.041
1.00 21.54
ATOM   1147  N   ASP 121      35.663  24.198  46.780
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ATOM   1149  CA  ASP 121      35.151  23.043  47.515
1.00 17.79
ATOM   1150  CB  ASP 121      34.292  23.489  48.709
1.00 16.14
ATOM   1151  CG  ASP 121      35.128  24.177  49.781
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ATOM   1152  OD1 ASP 121      36.279  23.733  49.983
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ATOM   1153  OD2 ASP  121      34.660  25.169  50.381
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ATOM   1154  C   ASP  121      34.431  22.078  46.565
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ATOM   1155  O   ASP  121      34.707  22.097  45.360
1.00 20.27
ATOM   1156  N   ARG  122      33.541  21.233  47.071
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ATOM   1158  CA  ARG  122      32.868  20.264  46.211
1.00 14.33
ATOM   1159  CB  ARG  122      33.468  18.871  46.417
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ATOM   1160  CG  ARG  122      34.987  18.856  46.370
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ATOM   1161  CD  ARG  122      35.555  17.493  46.647
1.00 25.64
ATOM   1162  NE  ARG  122      35.516  16.674  45.446
1.00 30.29
ATOM   1164  CZ  ARG  122      36.541  15.960  44.996
1.00 32.24
ATOM   1165  NH1 ARG  122      37.699  15.959  45.650
1.00 33.69
ATOM   1168  NH2 ARG  122      36.411  15.255  43.880
1.00 34.20
ATOM   1171  C   ARG  122      31.386  20.210  46.513
1.00 12.98
ATOM   1172  O   ARG  122      30.978  20.389  47.657
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ATOM   1173  N   THR  123      30.579  19.961  45.492
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ATOM   1175  CA  THR  123      29.144  19.878  45.705
1.00 11.52
ATOM   1176  CB  THR  123      28.517  21.287  45.856
1.00 12.34
ATOM   1177  OG1 THR  123      27.219  21.171  46.455
1.00 13.52
ATOM   1179  CG2 THR  123      28.408  22.004  44.503
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ATOM   1180  C   THR  123      28.507  19.108  44.558
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ATOM   1181  O   THR  123      29.212  18.577  43.694
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ATOM   1182  N   THR  124      27.187  18.988  44.592
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ATOM   1184  CA  THR  124      26.451  18.283  43.548
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ATOM   1185  CB  THR  124      25.470  17.273  44.172
1.00 11.44
ATOM   1186  OG1 THR  124      26.212  16.222  44.818
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ATOM   1188  CG2 THR  124      24.564  16.679  43.105
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ATOM   1189  C   THR  124      25.691  19.343  42.738
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ATOM   1190  O   THR  124      25.013  20.201  43.307
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ATOM   1193  CA  PHE 125      25.172  20.310  40.581
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ATOM   1194  CB  PHE 125      26.040  21.581  40.430
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ATOM   1195  CG  PHE 125      27.430  21.321  39.899
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ATOM   1196  CD1 PHE 125      28.486  21.072  40.771
1.00 10.51
ATOM   1197  CD2 PHE 125      27.681  21.298  38.526
1.00 10.72
ATOM   1198  CE1 PHE 125      29.760  20.801  40.290
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ATOM   1199  CE2 PHE 125      28.956  21.027  38.037
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ATOM   1200  CZ  PHE 125      29.991  20.779  38.911
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ATOM   1201  C   PHE 125      24.813  19.744  39.216
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ATOM   1202  O   PHE 125      25.350  18.720  38.791
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ATOM   1203  N   THR 126      23.886  20.418  38.547
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ATOM   1205  CA  THR 126      23.431  20.026  37.222
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ATOM   1206  CB  THR 126      21.949  20.388  37.041
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ATOM   1207  OG1 THR 126      21.192  19.813  38.113
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ATOM   1209  CG2 THR 126      21.416  19.847  35.715
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ATOM   1210  C   THR 126      24.223  20.729  36.122
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ATOM   1211  O   THR 126      24.701  21.857  36.305
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ATOM   1212  N   GLN 127      24.389  20.028  35.005
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ATOM   1214  CA  GLN 127      25.075  20.560  33.832
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ATOM   1215  CB  GLN 127      26.264  19.683  33.444
1.00  7.94
ATOM   1216  CG  GLN 127      27.314  19.489  34.527
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ATOM   1217  CD  GLN 127      28.410  18.547  34.080
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ATOM   1218  OE1 GLN 127      28.173  17.364  33.849
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ATOM   1219  NE2 GLN 127      29.599  19.072  33.903
1.00  9.24
ATOM   1222  C   GLN 127      24.082  20.546  32.671
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ATOM   1223  O   GLN 127      23.335  19.570  32.500
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ATOM   1224  N   TYR 128      24.050  21.637  31.915
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ATOM   1227  CB  TYR  128      22.446  23.076  30.678
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ATOM   1228  CG  TYR  128      21.337  23.250  31.683
1.00  7.55
ATOM   1229  CD1 TYR  128      20.883  22.185  32.458
1.00  8.48
ATOM   1230  CE1 TYR  128      19.866  22.370  33.389
1.00  9.54
ATOM   1231  CD2 TYR  128      20.748  24.495  31.859
1.00  8.26
ATOM   1232  CE2 TYR  128      19.737  24.686  32.784
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ATOM   1233  CZ  TYR  128      19.304  23.625  33.543
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ATOM   1234  OH  TYR  128      18.303  23.828  34.475
1.00 10.00
ATOM   1236  C   TYR  128      24.143  21.720  29.539
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ATOM   1237  O   TYR  128      25.161  22.422  29.550
1.00  7.10
ATOM   1238  N   TRP  129      23.786  20.956  28.511
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ATOM   1240  CA  TRP  129      24.626  20.849  27.320
1.00  6.02
ATOM   1241  CB  TRP  129      25.266  19.455  27.203
1.00  6.55
ATOM   1242  CG  TRP  129      26.025  18.930  28.374
1.00 76.122  1.00  9.09
ATOM   1211  O   THR  126      24.701  21.857  36.305
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ATOM   1212  N   GLN  127      24.389  20.028  35.005
1.00  8.52
ATOM   1214  CA  GLN  127      25.075  20.560  33.832
1.00  7.83
ATOM   1215  CB  GLN  127      26.264  19.683  33.444
1.00  7.94
ATOM   1216  CG  GLN  127      27.314  19.489  34.527
1.00  8.42
ATOM   1217  CD  GLN  127      28.410  18.547  34.080
1.00  9.41
ATOM   1218  OE1 GLN  127      28.173  17.364  33.849
1.00 11.46
ATOM   1219  NE2 GLN  127      29.599  19.072  33.903
1.00  9.24
ATOM   1222  C   GLN  127      24.082  20.546  32.671
1.00  8.00
ATOM   1223  O   GLN  127      23.335  19.570  32.500
1.00  9.01
ATOM   1224  N   TYR  128      24.050  21.637  31.915
1.00  7.47
ATOM   1226  CA  TYR  128      23.193  21.747  30.725
1.00  7.08
ATOM   1227  CB  TYR  128      22.446  23.076  30.678
1.00  7.17
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ATOM   1229  CD1 TYR 128      20.883  22.185  32.458
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ATOM   1230  CE1 TYR 128      19.866  22.370  33.389
1.00  9.54
ATOM   1231  CD2 TYR 128      20.748  24.495  31.859
1.00  8.26
ATOM   1232  CE2 TYR 128      19.737  24.686  32.784
1.00  8.94
ATOM   1233  CZ  TYR 128      19.304  23.625  33.543
1.00  8.68
ATOM   1234  OH  TYR 128      18.303  23.828  34.475
1.00 10.00
ATOM   1236  C   TYR 128      24.143  21.720  29.539
1.00  6.99
ATOM   1237  O   TYR 128      25.161  22.422  29.550
1.00  7.10
ATOM   1238  N   TRP 129      23.786  20.956  28.511
1.00  6.30
ATOM   1240  CA  TRP 129      24.626  20.849  27.320
1.00  6.02
ATOM   1241  CB  TRP 129      25.266  19.455  27.203
1.00  6.55
ATOM   1242  CG  TRP 129      26.025  18.930  28.374
1.00  7.49
ATOM   1243  CD2 TRP 129      27.451  18.787  28.477
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ATOM   1244  CE2 TRP 129      27.722  18.209  29.731
1.00  8.17
ATOM   1245  CE3 TRP 129      28.522  19.086  27.622
1.00  7.59
ATOM   1246  CD1 TRP 129      25.506  18.444  29.541
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ATOM   1247  NE1 TRP 129      26.517  18.009  30.362
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ATOM   1249  CZ2 TRP 129      29.028  17.921  30.156
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ATOM   1250  CZ3 TRP 129      29.806  18.797  28.044
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ATOM   1251  CH2 TRP 129      30.049  18.221  29.297
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ATOM   1252  C   TRP 129      23.879  20.976  26.010
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ATOM   1253  O   TRP 129      22.750  20.503  25.891
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ATOM   1254  N   SER 130      24.532  21.630  25.051
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ATOM   1256  CA  SER 130      24.101  21.652  23.655
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ATOM   1257  CB  SER 130      23.813  23.049  23.120
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ATOM   1258  OG  SER 130      22.470  23.416  23.404
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ATOM   1260  C   SER 130      25.385  21.110  23.022
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ATOM   1262  N   VAL 131      25.313  19.945  22.387
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ATOM   1264  CA  VAL 131      26.490  19.328  21.762
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ATOM   1265  CB  VAL 131      26.797  17.953  22.401
1.00  6.01
ATOM   1266  CG1 VAL 131      28.104  17.379  21.836
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ATOM   1267  CG2 VAL 131      26.910  18.089  23.913
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ATOM   1268  C   VAL 131      26.230  19.155  20.269
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ATOM   1269  O   VAL 131      25.270  18.491  19.868
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ATOM   1270  N   ARG 132      27.075  19.755  19.433
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ATOM   1272  CA  ARG 132      26.877  19.667  17.989
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ATOM   1273  CB  ARG 132      27.971  20.447  17.262
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ATOM   1274  CG  ARG 132      27.492  21.119  15.982
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ATOM   1275  CD  ARG 132      28.601  22.044  15.465
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ATOM   1276  NE  ARG 132      28.257  22.750  14.230
1.00 10.15
ATOM   1278  CZ  ARG 132      28.405  22.248  13.007
1.00  9.87
ATOM   1279  NH1 ARG 132      28.883  21.020  12.830
1.00  9.74
ATOM   1282  NH2 ARG 132      28.098  22.993  11.956
1.00 11.62
ATOM   1285  C   ARG 132      26.867  18.194  17.560
1.00  5.99
ATOM   1286  O   ARG 132      27.605  17.374  18.110
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ATOM   1287  N   GLN 133      26.022  17.849  16.590
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ATOM   1289  CA  GLN 133      25.941  16.457  16.156
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ATOM   1290  CB  GLN 133      24.677  16.213  15.328
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ATOM   1291  CG  GLN 133      23.402  16.324  16.174
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ATOM   1292  CD  GLN 133      22.125  15.987  15.413
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ATOM   1293  OE1 GLN 133      21.084  15.701  16.022
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ATOM   1294  NE2 GLN 133      22.193  16.014  14.095
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ATOM   1298  O   GLN 133      27.554  14.821  15.497
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ATOM   1302  CB  SER 134      28.839  16.670  12.467
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ATOM   1303  OG  SER 134      28.295  17.904  12.025
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ATOM   1305  C   SER 134      30.024  17.825  14.382
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ATOM   1306  O   SER 134      29.552  18.911  14.738
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ATOM   1307  N   LYS 135      31.336  17.588  14.353
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ATOM   1309  CA  LYS 135      32.288  18.633  14.722
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ATOM   1310  CB  LYS 135      33.733  18.098  14.730
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ATOM   1311  CG  LYS 135      33.968  16.992  15.737
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ATOM   1312  CD  LYS 135      35.408  16.520  15.771
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ATOM   1313  CE  LYS 135      35.552  15.304  16.668
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ATOM   1314  NZ  LYS 135      36.989  14.926  16.843
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ATOM   1318  C   LYS 135      32.217  19.823  13.783
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ATOM   1319  O   LYS 135      32.134  19.664  12.554
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ATOM   1320  N   ARG 136      32.239  21.023  14.353
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ATOM   1322  CA  ARG 136      32.227  22.225  13.548
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ATOM   1323  CB  ARG 136      31.842  23.431  14.397
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ATOM   1324  CG  ARG 136      31.469  24.661  13.597
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ATOM   1325  CD  ARG 136      30.960  25.755  14.519
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ATOM   1326  NE  ARG 136      30.367  26.877  13.797
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ATOM   1328  CZ  ARG 136      29.614  27.817  14.361
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ATOM   1329  NH1 ARG 136      29.354  27.777  15.661
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ATOM   1332  NH2 ARG 136      29.121  28.807  13.631
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ATOM   1335  C   ARG 136      33.650  22.412  13.027
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ATOM   1336  O   ARG 136      34.609  22.278  13.785
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ATOM   1337  N   PRO 137      33.810  22.679  11.720
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ATOM   1338  CD  PRO 137      32.814  22.730  10.637
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ATOM   1340  CB  PRO  137      34.916  23.225   9.728
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ATOM   1341  CG  PRO  137      33.665  22.488   9.420
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ATOM   1342  C   PRO  137      35.810  24.072  11.917
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ATOM   1343  O   PRO  137      35.110  25.007  12.320
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ATOM   1344  N   THR  138      37.114  24.023  12.139
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ATOM   1346  CA  THR  138      37.795  25.116  12.811
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ATOM   1347  CB  THR  138      38.652  24.604  13.982
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ATOM   1348  OG1 THR  138      39.583  23.616  13.512
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ATOM   1350  CG2 THR  138      37.746  24.000  15.070
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ATOM   1351  C   THR  138      38.650  25.904  11.826
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ATOM   1352  O   THR  138      38.816  25.493  10.675
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ATOM   1353  N   GLY  139      39.132  27.061  12.259
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ATOM   1355  CA  GLY  139      39.954  27.889  11.397
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ATOM   1356  C   GLY  139      39.157  29.004  10.743
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ATOM   1357  O   GLY  139      39.730  29.873  10.085
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ATOM   1358  N   SER  140      37.839  28.981  10.930
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ATOM   1360  CA  SER  140      36.954  29.996  10.375
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ATOM   1361  CB  SER  140      35.857  29.343   9.538
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ATOM   1362  OG  SER  140      36.404  28.701   8.403
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ATOM   1364  C   SER  140      36.293  30.823  11.468
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ATOM   1365  O   SER  140      36.369  30.485  12.657
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ATOM   1366  N   ASN  141      35.697  31.937  11.064
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ATOM   1368  CA  ASN  141      34.988  32.815  11.988
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ATOM   1369  CB  ASN  141      34.774  34.193  11.357
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ATOM   1370  CG  ASN  141      36.068  34.965  11.187
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ATOM   1371  OD1 ASN  141      37.056  34.714  11.885
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ATOM   1372  ND2 ASN  141      36.066  35.914  10.270
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ATOM   1376  O   ASN 141      32.787  32.050  11.399
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ATOM   1377  N   ALA 142      33.477  31.712  13.523
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ATOM   1379  CA  ALA 142      32.265  31.022  13.945
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ATOM   1380  CB  ALA 142      32.629  29.707  14.600
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ATOM   1381  C   ALA 142      31.457  31.875  14.903
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ATOM   1382  O   ALA 142      32.001  32.719  15.618
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ATOM   1383  N   THR 143      30.155  31.627  14.927
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ATOM   1385  CA  THR 143      29.246  32.357  15.788
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ATOM   1386  CB  THR 143      28.277  33.232  14.953
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ATOM   1387  OG1 THR 143      29.033  34.077  14.069
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ATOM   1389  CG2 THR 143      27.412  34.092  15.847
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ATOM   1390  C   THR 143      28.389  31.394  16.596
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ATOM   1391  O   THR 143      28.011  30.319  16.114
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ATOM   1392  N   ILE 144      28.179  31.740  17.859
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ATOM   1394  CA  ILE 144      27.297  30.977  18.725
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ATOM   1395  CB  ILE 144      28.030  30.341  19.930
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ATOM   1396  CG2 ILE 144      26.998  29.785  20.946
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ATOM   1397  CG1 ILE 144      28.951  29.218  19.445
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ATOM   1398  CD1 ILE 144      29.793  28.590  20.549
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ATOM   1399  C   ILE 144      26.262  31.992  19.213
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ATOM   1400  O   ILE 144      26.592  32.926  19.951
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ATOM   1401  N   THR 145      25.033  31.857  18.721
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ATOM   1403  CA  THR 145      23.929  32.739  19.102
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ATOM   1404  CB  THR 145      22.930  32.837  17.950
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ATOM   1405  OG1 THR 145      23.616  33.323  16.786
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ATOM   1407  CG2 THR 145      21.813  33.805  18.295
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ATOM   1408  C   THR 145      23.331  32.077  20.345
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ATOM   1409  O   THR 145      22.444  31.214  20.262
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ATOM   1410  N   PHE 146      23.861  32.470  21.500
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ATOM   1412  CA  PHE 146      23.486  31.856  22.761
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ATOM   1413  CB  PHE 146      24.361  32.397  23.893
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ATOM   1414  CG  PHE 146      24.232  31.635  25.189
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ATOM   1415  CD1 PHE 146      24.328  30.247  25.221
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ATOM   1416  CD2 PHE 146      24.032  32.318  26.380
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ATOM   1417  CE1 PHE 146      24.231  29.560  26.429
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ATOM   1418  CE2 PHE 146      23.934  31.636  27.589
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ATOM   1419  CZ  PHE 146      24.034  30.259  27.611
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ATOM   1420  C   PHE 146      22.016  31.897  23.106
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ATOM   1421  O   PHE 146      21.493  30.937  23.682
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ATOM   1422  N   THR 147      21.330  32.950  22.677
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ATOM   1424  CA  THR 147      19.907  33.059  22.958
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ATOM   1425  CB  THR 147      19.331  34.382  22.419
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ATOM   1426  OG1 THR 147      17.947  34.473  22.777
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ATOM   1428  CG2 THR 147      19.433  34.429  20.930
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ATOM   1429  C   THR 147      19.109  31.868  22.396
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ATOM   1430  O   THR 147      18.140  31.429  23.017
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ATOM   1431  N   ASN 148      19.526  31.325  21.250
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ATOM   1433  CA  ASN 148      18.829  30.176  20.655
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ATOM   1434  CB  ASN 148      19.423  29.784  19.302
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ATOM   1435  CG  ASN 148      19.127  30.791  18.202
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ATOM   1436  OD1 ASN 148      19.860  30.861  17.201
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ATOM   1437  ND2 ASN 148      18.069  31.569  18.367
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ATOM   1440  C   ASN 148      18.909  28.967  21.574
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ATOM   1441  O   ASN 148      17.949  28.202  21.697
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ATOM   1442  N   HIS 149      20.056  28.804  22.232
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ATOM 1444 CA HIS 149 20.264 27.693 23.149 1.00 7.07
ATOM 1445 CB HIS 149 21.755 27.507 23.426 1.00 6.26
ATOM 1446 CG HIS 149 22.521 27.095 22.208 1.00 6.45
ATOM 1447 CD2 HIS 149 23.157 27.828 21.262 1.00 7.33
ATOM 1448 ND1 HIS 149 22.654 25.776 21.826 1.00 7.11
ATOM 1450 CE1 HIS 149 23.339 25.716 20.697 1.00 7.85
ATOM 1451 NE2 HIS 149 23.655 26.947 20.335 1.00 7.63
ATOM 1453 C HIS 149 19.477 27.883 24.430 1.00 7.20
ATOM 1454 O HIS 149 18.780 26.971 24.863 1.00 8.12
ATOM 1455 N VAL 150 19.552 29.080 25.005 1.00 7.02
ATOM 1457 CA VAL 150 18.813 29.393 26.223 1.00 7.84
ATOM 1458 CB VAL 150 19.044 30.858 26.632 1.00 7.77
ATOM 1459 CG1 VAL 150 18.131 31.239 27.801 1.00 8.91
ATOM 1460 CG2 VAL 150 20.515 31.052 27.014 1.00 8.40
ATOM 1461 C VAL 150 17.313 29.139 26.017 1.00 8.70
ATOM 1462 O VAL 150 16.661 28.489 26.848 1.00 9.04
ATOM 1463 N ASN 151 16.794 29.575 24.875 1.00 9.29
ATOM 1465 CA ASN 151 15.379 29.396 24.577 1.00 11.27
ATOM 1466 CB ASN 151 14.967 30.276 23.397 1.00 13.99
ATOM 1467 CG ASN 151 14.954 31.754 23.767 1.00 17.23
ATOM 1468 OD1 ASN 151 14.736 32.099 24.932 1.00 22.12
ATOM 1469 ND2 ASN 151 15.210 32.624 22.797 1.00 19.31
ATOM 1472 C ASN 151 14.993 27.933 24.372 1.00 10.65
ATOM 1473 O ASN 151 13.957 27.484 24.875 1.00 11.12
ATOM 1474 N ALA 152 15.828 27.180 23.660 1.00 9.31
ATOM 1476 CA ALA 152 15.563 25.754 23.459 1.00 8.90
ATOM 1477 CB ALA 152 16.560 25.156 22.476 1.00 10.29
ATOM 1478 C ALA 152 15.629 25.011 24.797 1.00 9.31

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ATOM   1479  O   ALA  152      14.812  24.121  25.066
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ATOM   1480  N   TRP  153      16.589  25.372  25.642
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ATOM   1482  CA  TRP  153      16.723  24.728  26.941
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ATOM   1483  CB  TRP  153      17.970  25.239  27.677
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ATOM   1484  CG  TRP  153      19.290  24.835  27.040
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ATOM   1485  CD2 TRP  153      20.590  25.321  27.401
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ATOM   1486  CE2 TRP  153      21.528  24.676  26.560
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ATOM   1487  CE3 TRP  153      21.054  26.243  28.350
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ATOM   1488  CD1 TRP  153      19.480  23.940  26.022
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ATOM   1489  NE1 TRP  153      20.829  23.836  25.729
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ATOM   1491  CZ2 TRP  153      22.905  24.925  26.642
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ATOM   1492  CZ3 TRP  153      22.420  26.491  28.429
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ATOM   1493  CH2 TRP  153      23.327  25.834  27.581
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ATOM   1494  C   TRP  153      15.479  25.002  27.787
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ATOM   1495  O   TRP  153      14.936  24.096  28.422
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ATOM   1496  N   LYS  154      15.012  26.245  27.753
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ATOM   1498  CA  LYS  154      13.834  26.636  28.514
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ATOM   1499  CB  LYS  154      13.571  28.129  28.322
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ATOM   1500  CG  LYS  154      12.405  28.650  29.100
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ATOM   1501  CD  LYS  154      12.336  30.169  29.040
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ATOM   1502  CE  LYS  154      11.104  30.667  29.794
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ATOM   1503  NZ  LYS  154      11.069  30.150  31.208
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ATOM   1507  C   LYS  154      12.620  25.803  28.091
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ATOM   1508  O   LYS  154      11.847  25.360  28.938
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ATOM   1509  N   SER  155      12.499  25.520  26.797
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ATOM   1511  CA  SER  155      11.372  24.732  26.298
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ATOM   1512  CB  SER  155      11.358  24.706  24.770
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ATOM   1513  OG  SER  155      12.315  23.786  24.267
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ATOM   1517  N   HIS 156      12.530  22.847  27.316
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ATOM   1519  CA  HIS 156      12.653  21.502  27.865
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ATOM   1520  CB  HIS 156      13.846  20.777  27.243
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ATOM   1521  CG  HIS 156      13.660  20.483  25.795
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ATOM   1522  CD2 HIS 156      12.918  19.540  25.170
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ATOM   1523  ND1 HIS 156      14.220  21.251  24.799
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ATOM   1525  CE1 HIS 156      13.832  20.795  23.622
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ATOM   1530  N   GLY 157      12.349  22.607  29.991
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ATOM   1532  CA  GLY 157      12.404  22.719  31.438
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ATOM   1533  C   GLY 157      13.801  22.895  32.022
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ATOM   1534  O   GLY 157      14.016  22.658  33.204
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ATOM   1535  N   MET 158      14.746  23.333  31.202
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ATOM   1537  CA  MET 158      16.114  23.548  31.651
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ATOM   1538  CB  MET 158      17.083  23.009  30.598
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ATOM   1539  CG  MET 158      17.075  21.493  30.479
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ATOM   1540  SD  MET 158      17.768  20.929  28.919
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ATOM   1541  CE  MET 158      19.481  21.502  29.063
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ATOM   1542  C   MET 158      16.297  25.047  31.839
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ATOM   1543  O   MET 158      16.681  25.756  30.915
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ATOM   1544  N   ASN 159      15.965  25.529  33.029
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ATOM   1546  CA  ASN 159      16.067  26.948  33.337
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ATOM   1547  CB  ASN 159      14.883  27.387  34.197
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ATOM   1548  CG  ASN 159      13.557  27.198  33.486
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ATOM   1549  OD1 ASN   159      12.757  26.340  33.857
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ATOM   1550  ND2 ASN   159      13.344  27.961  32.425
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ATOM   1553  C   ASN   159      17.367  27.289  34.039
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ATOM   1554  O   ASN   159      17.803  26.584  34.955
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ATOM   1555  N   LEU   160      17.982  28.378  33.603
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ATOM   1557  CA  LEU   160      19.239  28.825  34.178
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ATOM   1558  CB  LEU   160      19.997  29.674  33.152
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ATOM   1559  CG  LEU   160      20.564  28.838  32.002
1.00 13.30
ATOM   1560  CD1 LEU   160      20.954  29.739  30.855
1.00 14.40
ATOM   1561  CD2 LEU   160      21.758  28.020  32.482
1.00 13.55
ATOM   1562  C   LEU   160      19.007  29.596  35.480
1.00 14.33
ATOM   1563  O   LEU   160      17.882  30.024  35.763
1.00 14.67
ATOM   1564  N   GLY   161      20.060  29.740  36.278
1.00 14.47
ATOM   1566  CA  GLY   161      19.953  30.465  37.529
1.00 15.47
ATOM   1567  C   GLY   161      19.784  31.954  37.290
1.00 16.15
ATOM   1568  O   GLY   161      19.999  32.452  36.171
1.00 16.60
ATOM   1569  N   SER   162      19.445  32.683  38.347
1.00 17.37
ATOM   1571  CA  SER   162      19.239  34.121  38.230
1.00 17.84
ATOM   1572  CB  SER   162      18.170  34.576  39.223
1.00 19.00
ATOM   1573  OG  SER   162      18.500  34.145  40.530
1.00 21.73
ATOM   1575  C   SER   162      20.505  34.953  38.413
1.00 17.19
ATOM   1576  O   SER   162      20.538  36.123  38.041
1.00 17.92
ATOM   1577  N   ASN   163      21.541  34.362  38.995
1.00 16.21
ATOM   1579  CA  ASN   163      22.784  35.087  39.219
1.00 15.87
ATOM   1580  CB  ASN   163      23.260  34.871  40.661
1.00 19.24
ATOM   1581  CG  ASN   163      24.477  35.718  41.020
1.00 23.45
ATOM   1582  OD1 ASN   163      25.295  35.326  41.860
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ATOM   1583  ND2 ASN   163      24.590  36.894  40.410
1.00 25.66
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ATOM   1590  CA  TRP  164      25.106  35.017  36.205
1.00 12.67
ATOM   1591  CB  TRP  164      24.802  35.764   9.997
29.674  33.152  1.00 13.40
ATOM   1559  CG  LEU  160      20.564  28.838  32.002
1.00 13.30
ATOM   1560  CD1 LEU  160      20.954  29.739  30.855
1.00 14.40
ATOM   1561  CD2 LEU  160      21.758  28.020  32.482
1.00 13.55
ATOM   1562  C   LEU  160      19.007  29.596  35.480
1.00 14.33
ATOM   1563  O   LEU  160      17.882  30.024  35.763
1.00 14.67
ATOM   1564  N   GLY  161      20.060  29.740  36.278
1.00 14.47
ATOM   1566  CA  GLY  161      19.953  30.465  37.529
1.00 15.47
ATOM   1567  C   GLY  161      19.784  31.954  37.290
1.00 16.15
ATOM   1568  O   GLY  161      19.999  32.452  36.171
1.00 16.60
ATOM   1569  N   SER  162      19.445  32.683  38.347
1.00 17.37
ATOM   1571  CA  SER  162      19.239  34.121  38.230
1.00 17.84
ATOM   1572  CB  SER  162      18.170  34.576  39.223
1.00 19.00
ATOM   1573  OG  SER  162      18.500  34.145  40.530
1.00 21.73
ATOM   1575  C   SER  162      20.505  34.953  38.413
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ATOM   1576  O   SER  162      20.538  36.123  38.041
1.00 17.92
ATOM   1577  N   ASN  163      21.541  34.362  38.995
1.00 16.21
ATOM   1579  CA  ASN  163      22.784  35.087  39.219
1.00 15.87
ATOM   1580  CB  ASN  163      23.260  34.871  40.661
1.00 19.24
ATOM   1581  CG  ASN  163      24.477  35.718  41.020
1.00 23.45
ATOM   1582  OD1 ASN  163      25.295  35.326  41.860
1.00 27.11
ATOM   1583  ND2 ASN  163      24.590  36.894  40.410
1.00 25.66
ATOM   1586  C   ASN  163      23.816  34.580  38.219
1.00 14.67
ATOM   1587  O   ASN  163      24.280  33.454  38.320
1.00 15.00
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ATOM   1591  CB  TRP 164      24.802  35.764  34.902
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ATOM   1592  CG  TRP 164      23.523  35.331  34.250
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ATOM   1593  CD2 TRP 164      23.393  34.489  33.098
1.00 13.58
ATOM   1594  CE2 TRP 164      22.015  34.297  32.871
1.00 14.09
ATOM   1595  CE3 TRP 164      24.308  33.876  32.238
1.00 13.32
ATOM   1596  CD1 TRP 164      22.253  35.620  34.659
1.00 15.26
ATOM   1597  NE1 TRP 164      21.340  34.996  33.837
1.00 15.80
ATOM   1599  CZ2 TRP 164      21.534  33.518  31.825
1.00 14.29
ATOM   1600  CZ3 TRP 164      23.829  33.107  31.204
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ATOM   1601  CH2 TRP 164      22.456  32.932  31.004
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ATOM   1602  C   TRP 164      26.576  35.218  36.569
1.00 12.17
ATOM   1603  O   TRP 164      26.990  36.300  37.001
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ATOM   1604  N   ALA 165      27.365  34.173  36.360
1.00 10.88
ATOM   1606  CA  ALA 165      28.796  34.233  36.611
1.00  9.93
ATOM   1607  CB  ALA 165      29.275  32.979  37.333
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ATOM   1608  C   ALA 165      29.486  34.396  35.246
1.00  9.94
ATOM   1609  O   ALA 165      28.846  34.822  34.275
1.00 10.07
ATOM   1610  N   TYR 166      30.759  34.027  35.145
1.00  8.78
ATOM   1612  CA  TYR 166      31.474  34.207  33.881
1.00  8.27
ATOM   1613  CB  TYR 166      32.982  34.002  34.069
1.00  8.85
ATOM   1614  CG  TYR 166      33.384  32.610  34.503
1.00 10.16
ATOM   1615  CD1 TYR 166      33.438  31.567  33.588
1.00 10.41
ATOM   1616  CE1 TYR 166      33.821  30.302  33.964
1.00 12.25
ATOM   1617  CD2 TYR 166      33.726  32.344  35.827
1.00  9.77
ATOM   1618  CE2 TYR 166      34.111  31.073  36.220
1.00 11.27
ATOM   1619  CZ  TYR 166      34.157  30.061  35.283
1.00 11.58
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ATOM	1620	OH	TYR	166	34.553	28.803	35.642
1.00	13.25						
ATOM	1622	C	TYR	166	30.941	33.364	32.717
1.00	8.20						
ATOM	1623	O	TYR	166	30.242	32.355	32.915
1.00	8.68						
ATOM	1624	N	GLN	167	31.327	33.765	31.509
1.00	8.01						
ATOM	1626	CA	GLN	167	30.894	33.122	30.272
1.00	7.76						
ATOM	1627	CB	GLN	167	29.687	33.897	29.742
1.00	8.57						
ATOM	1628	CG	GLN	167	29.059	33.379	28.465
1.00	9.03						
ATOM	1629	CD	GLN	167	27.822	34.189	28.116
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ATOM	1630	OE1	GLN	167	27.896	35.405	27.941
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ATOM	1631	NE2	GLN	167	26.674	33.531	28.081
1.00	11.48						
ATOM	1634	C	GLN	167	32.084	33.233	29.322
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ATOM	1635	O	GLN	167	32.463	34.340	28.940
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ATOM	1636	N	VAL	168	32.684	32.103	28.961
1.00	6.64						
ATOM	1638	CA	VAL	168	33.871	32.126	28.109
1.00	6.81						
ATOM	1639	CB	VAL	168	35.152	31.861	28.970
1.00	7.79						
ATOM	1640	CG1	VAL	168	35.294	32.912	30.062
1.00	7.77						
ATOM	1641	CG2	VAL	168	35.093	30.475	29.599
1.00	8.95						
ATOM	1642	C	VAL	168	33.857	31.097	26.992
1.00	6.72						
ATOM	1643	O	VAL	168	33.087	30.141	27.034
1.00	7.62						
ATOM	1644	N	MET	169	34.671	31.325	25.962
1.00	6.24						
ATOM	1646	CA	MET	169	34.822	30.353	24.875
1.00	7.10						
ATOM	1647	CB	MET	169	35.078	31.020	23.527
1.00	7.60						
ATOM	1648	CG	MET	169	35.016	30.028	22.389
1.00	9.98						
ATOM	1649	SD	MET	169	33.355	29.295	22.243
1.00	10.60						
ATOM	1650	CE	MET	169	33.759	27.805	21.363
1.00	11.26						
ATOM	1651	C	MET	169	36.066	29.618	25.357
1.00	7.23						
ATOM	1652	O	MET	169	37.175	30.167	25.347
1.00	7.62						
ATOM	1653	N	ALA	170	35.871	28.375	25.750
1.00	6.93						

ATOM	1655	CA	ALA	170	36.920	27.585	26.367
1.00	7.05						
ATOM	1656	CB	ALA	170	36.453	27.205	27.778
1.00	8.93						
ATOM	1657	C	ALA	170	37.436	26.334	25.706
1.00	7.41						
ATOM	1658	O	ALA	170	36.823	25.752	24.806
1.00	6.87						
ATOM	1659	N	THR	171	38.585	25.920	26.230
1.00	7.05						
ATOM	1661	CA	THR	171	39.218	24.668	25.892
1.00	6.62						
ATOM	1662	CB	THR	171	40.690	24.847	25.497
1.00	7.70						
ATOM	1663	OG1	THR	171	40.771	25.554	24.258
1.00	8.40						
ATOM	1665	CG2	THR	171	41.363	23.476	25.341
1.00	8.06						
ATOM	1666	C	THR	171	39.195	23.916	27.237
1.00	7.48						
ATOM	1667	O	THR	171	39.717	24.420	28.242
1.00	7.78						
ATOM	1668	N	GLU	172	38.507	22.778	27.290
1.00	7.60						
ATOM	1670	CA	GLU	172	38.473	21.968	28.510
1.00	7.72						
ATOM	1671	CB	GLU	172	37.040	21.751	29.035
1.00	8.64						
ATOM	1672	CG	GLU	172	37.010	20.705	30.165
1.00	10.82						
ATOM	1673	CD	GLU	172	35.722	20.667	30.977
1.00	13.27						
ATOM	1674	OE1	GLU	172	35.663	19.872	31.946
1.00	14.50						
ATOM	1675	OE2	GLU	172	34.785	21.428	30.667
1.00	13.18						
ATOM	1676	C	GLU	172	39.085	20.613	28.180
1.00	8.52						
ATOM	1677	O	GLU	172	38.843	20.069	27.101
1.00	9.45						
ATOM	1678	N	GLY	173	39.909	20.089	29.077
1.00	7.84						
ATOM	1680	CA	GLY	173	40.501	18.792	28.830
1.00	8.42						
ATOM	1681	C	GLY	173	40.389	17.910	30.046
1.00	9.01						
ATOM	1682	O	GLY	173	40.196	18.403	31.164
1.00	9.09						
ATOM	1683	N	TYR	174	40.442	16.602	29.817
1.00	9.90						
ATOM	1685	CA	TYR	174	40.389	15.626	30.896
1.00	10.77						
ATOM	1686	CB	TYR	174	38.969	15.103	31.125
1.00	13.07						
ATOM	1687	CG	TYR	174	38.914	14.153	32.296
1.00	16.61						

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ATOM   1688  CD1 TYR   174      38.603  12.811  32.115
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ATOM   1689  CE1 TYR   174      38.619  11.926  33.181
1.00 19.66
ATOM   1690  CD2 TYR   174      39.236  14.586  33.579
1.00 18.49
ATOM   1691  CE2 TYR   174      39.258  13.709  34.648
1.00 20.51
ATOM   1692  CZ  TYR   174      38.948  12.383  34.443
1.00 20.34
ATOM   1693  OH  TYR   174      38.970  11.506  35.508
1.00 24.58
ATOM   1695  C   TYR   174      41.311  14.466  30.552
1.00 10.26
ATOM   1696  O   TYR   174      41.107  13.773  29.553
1.00 10.33
ATOM   1697  N   GLN   175      42.359  14.308  31.359
1.00  9.98
ATOM   1699  CA  GLN   175      43.354  13.248  31.184
1.00 10.93
ATOM   1700  CB  GLN   175      42.837  11.937  31.787
1.00 13.41
ATOM   1701  CG  GLN   175      42.566  12.065  33.283
1.00 16.14
ATOM   1702  CD  GLN   175      42.419  10.732  34.001
1.00 19.68
ATOM   1703  OE1 GLN   175      42.573  10.665  35.228
1.00 21.27
ATOM   1704  NE2 GLN   175      42.110   9.673  33.258
1.00 20.43
ATOM   1707  C   GLN   175      43.781  13.073  29.720
1.00 10.40
ATOM   1708  O   GLN   175      43.726  11.976  29.151
1.00 10.98
ATOM   1709  N   SER   176      44.279  14.155  29.137
1.00 10.43
ATOM   1711  CA  SER   176      44.680  14.133  27.746
1.00  9.15
ATOM   1712  CB  SER   176      43.467  14.513  26.879
1.00 10.20
ATOM   1713  OG  SER   176      43.042  15.847  27.130
1.00  9.79
ATOM   1715  C   SER   176      45.840  15.101  27.489
1.00  9.26
ATOM   1716  O   SER   176      46.427  15.654  28.426
1.00  9.17
ATOM   1717  N   SER   177      46.178  15.264  26.216
1.00  9.64
ATOM   1719  CA  SER   177      47.240  16.164  25.776
1.00  9.15
ATOM   1720  CB  SER   177      48.507  15.375  25.461
1.00 10.09
ATOM   1721  OG  SER   177      48.928  14.633  26.596
1.00 12.00
ATOM   1723  C   SER   177      46.745  16.829  24.508
1.00 10.02
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ATOM   1724  O   SER 177      45.803  16.341  23.883
1.00  9.82
ATOM   1725  N   GLY 178      47.317  17.972  24.157
1.00  9.77
ATOM   1727  CA  GLY 178      46.893  18.632  22.935
1.00  9.38
ATOM   1728  C   GLY 178      47.289  20.089  22.869
1.00  9.30
ATOM   1729  O   GLY 178      48.044  20.569  23.714
1.00  8.99
ATOM   1730  N   SER 179      46.757  20.785  21.870
1.00  9.17
ATOM   1732  CA  SER 179      47.028  22.206  21.674
1.00  9.44
ATOM   1733  CB  SER 179      48.177  22.408  20.671
1.00 11.55
ATOM   1734  OG  SER 179      49.424  22.073  21.243
1.00 19.58
ATOM   1736  C   SER 179      45.804  22.901  21.104
1.00  8.63
ATOM   1737  O   SER 179      45.009  22.284  20.394
1.00  9.53
ATOM   1738  N   SER 180      45.636  24.164  21.463
1.00  8.95
ATOM   1740  CA  SER 180      44.563  24.989  20.937
1.00  8.51
ATOM   1741  CB  SER 180      43.263  24.866  21.750
1.00  9.65
ATOM   1742  OG  SER 180      43.365  25.508  23.006
1.00 11.87
ATOM   1744  C   SER 180      45.026  26.441  20.912
1.00  8.80
ATOM   1745  O   SER 180      45.920  26.848  21.674
1.00  9.51
ATOM   1746  N   ASN 181      44.454  27.202  19.993
1.00  8.43
ATOM   1748  CA  ASN 181      44.750  28.619  19.850
1.00  8.44
ATOM   1749  CB  ASN 181      45.837  28.845  18.792
1.00  9.20
ATOM   1750  CG  ASN 181      46.374  30.257  18.808
1.00 10.45
ATOM   1751  OD1 ASN 181      47.577  30.476  19.001
1.00 14.84
ATOM   1752  ND2 ASN 181      45.504  31.221  18.607
1.00  9.73
ATOM   1755  C   ASN 181      43.406  29.157  19.385
1.00  8.65
ATOM   1756  O   ASN 181      42.912  28.774  18.322
1.00  8.58
ATOM   1757  N   VAL 182      42.802  30.003  20.211
1.00  7.83
ATOM   1759  CA  VAL 182      41.474  30.535  19.940
1.00  7.20
ATOM   1760  CB  VAL 182      40.449  29.879  20.899
1.00  8.34
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ATOM   1761  CG1 VAL   182      39.018  30.269  20.525
1.00  9.23
ATOM   1762  CG2 VAL   182      40.612  28.362  20.910
1.00  8.07
ATOM   1763  C   VAL   182      41.411  32.046  20.158
1.00  8.01
ATOM   1764  O   VAL   182      42.013  32.576  21.097
1.00  7.62
ATOM   1765  N   THR   183      40.646  32.728  19.311
1.00  7.00
ATOM   1767  CA  THR   183      40.459  34.173  19.418
1.00  7.39
ATOM   1768  CB  THR   183      40.973  34.877  18.146
1.00  8.24
ATOM   1769  OG1 THR   183      42.362  34.565  17.995
1.00  9.42
ATOM   1771  CG2 THR   183      40.799  36.398  18.245
1.00  8.99
ATOM   1772  C   THR   183      38.964  34.373  19.603
1.00  7.65
ATOM   1773  O   THR   183      38.166  33.828  18.834
1.00  7.43
ATOM   1774  N   VAL   184      38.595  35.121  20.634
1.00  7.62
ATOM   1776  CA  VAL   184      37.194  35.351  20.999
1.00  8.26
ATOM   1777  CB  VAL   184      36.979  34.886  22.464
1.00  9.19
ATOM   1778  CG1 VAL   184      35.502  34.914  22.837
1.00  9.70
ATOM   1779  CG2 VAL   184      37.576  33.502  22.672
1.00  9.78
ATOM   1780  C   VAL   184      36.829  36.832  20.938
1.00  8.76
ATOM   1781  O   VAL   184      37.637  37.674  21.330
1.00  8.92
ATOM   1782  N   TRP   185      35.619  37.144  20.461
1.00  8.50
ATOM   1784  CA  TRP   185      35.142  38.525  20.401
1.00  9.38
ATOM   1785  CB  TRP   185      35.755  39.295  19.212
1.00  9.55
ATOM   1786  CG  TRP   185      35.302  38.875  17.840
1.00 10.25
ATOM   1787  CD2 TRP   185      35.814  37.791  17.055
1.00 10.55
ATOM   1788  CE2 TRP   185      35.150  37.825  15.808
1.00 10.75
ATOM   1789  CE3 TRP   185      36.769  36.797  17.281
1.00 10.39
ATOM   1790  CD1 TRP   185      34.367  39.497  17.061
1.00 11.62
ATOM   1791  NE1 TRP   185      34.272  38.876  15.840
1.00 12.03
ATOM   1793  CZ2 TRP   185      35.410  36.903  14.794
1.00 11.32
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ATOM	1794	CZ3	TRP	185	37.025	35.883	16.266
1.00	11.87						
ATOM	1795	CH2	TRP	185	36.346	35.944	15.040
1.00	11.03						
ATOM	1796	C	TRP	185	33.611	38.581	20.364
1.00	9.55						
ATOM	1797	O	TRP	185	32.974	37.509	20.372
1.00	9.75						
ATOM	1798	OXT	TRP	185	33.062	39.698	20.362
1.00	12.79						

The three-dimensional crystal structure of the *T. harzianum* xylanase shows a great degree of similarity to the structure of the *B. circulans* xylanase (Figure 3), although the *T. harzianum* xylanase contains two extra strands at the beginning of sheets I and II, and a few small insertions and deletions (see also Figure 1). While the structures are similar at 89% of the residues of the *B. circulans* xylanase, the sequences are identical at only 51% of the 185 residues. The x-ray co-ordinates of the structure is given below.

ATOM	1	CB	GLN	1	15.828	12.804	3.943
1.00	26.76						
ATOM	2	CG	GLN	1	14.520	12.576	3.185
1.00	31.36						
ATOM	3	CD	GLN	1	13.427	13.621	3.366
1.00	32.78						
ATOM	4	OE1	GLN	1	13.524	14.505	4.210
1.00	35.55						
ATOM	5	NE2	GLN	1	12.296	13.583	2.708
1.00	35.11						
ATOM	6	C	GLN	1	17.269	13.179	5.908
1.00	24.81						
ATOM	7	O	GLN	1	18.126	13.319	5.064
1.00	27.64						
ATOM	8	N	GLN	1	15.208	13.707	6.190
1.00	24.75						
ATOM	9	CA	GLN	1	15.926	12.688	5.486
1.00	25.70						
ATOM	10	N	THR	2	17.472	13.314	7.213
1.00	25.76						
ATOM	11	CA	THR	2	18.554	14.044	7.875
1.00	24.44						
ATOM	12	CB	THR	2	19.855	13.864	7.084
1.00	25.32						
ATOM	13	OG1	THR	2	20.017	12.431	7.146
1.00	26.72						

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ATOM     14  CG2 THR     2      21.104  14.549   7.600
1.00 27.32
ATOM     15  C   THR     2      17.847  15.388   7.733
1.00 21.71
ATOM     16  O   THR     2      17.768  16.039   6.692
1.00 20.85
ATOM     17  N   ILE     3      17.002  15.542   8.751
1.00 18.32
ATOM     18  CA  ILE     3      16.196  16.735   8.873
1.00 17.14
ATOM     19  CB  ILE     3      14.723  16.216   8.640
1.00 17.94
ATOM     20  CG2 ILE     3      13.680  16.289   9.744
1.00 16.71
ATOM     21  CG1 ILE     3      14.269  17.095   7.557
1.00 18.00
ATOM     22  CD1 ILE     3      14.803  16.716   6.179
1.00 20.15
ATOM     23  C   ILE     3      16.568  17.281  10.229
1.00 16.13
ATOM     24  O   ILE     3      17.085  16.584  11.119
1.00 16.34
ATOM     25  N   GLY     4      16.451  18.588  10.370
1.00 14.88
ATOM     26  CA  GLY     4      16.674  19.198  11.659
1.00 11.55
ATOM     27  C   GLY     4      15.299  19.349  12.251
1.00 12.16
ATOM     28  O   GLY     4      14.314  18.938  11.628
1.00 11.77
ATOM     29  N   PRO     5      15.134  19.926  13.445
1.00 11.94
ATOM     30  CD  PRO     5      16.159  20.593  14.231
1.00 12.27
ATOM     31  CA  PRO     5      13.887  19.803  14.160
1.00 12.67
ATOM     32  CB  PRO     5      14.208  20.405  15.490
1.00 12.77
ATOM     33  CG  PRO     5      15.703  20.207  15.627
1.00 12.89
ATOM     34  C   PRO     5      12.754  20.463  13.391
1.00 13.39
ATOM     35  O   PRO     5      12.974  21.493  12.764
1.00 14.17
ATOM     36  N   GLY     6      11.583  19.867  13.377
1.00 12.31
ATOM     37  CA  GLY     6      10.494  20.387  12.606
1.00 13.71
ATOM     38  C   GLY     6       9.260  19.487  12.636
1.00 14.99
ATOM     39  O   GLY     6       9.172  18.391  13.233
1.00 14.55
ATOM     40  N   THR     7       8.292  19.986  11.875
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ATOM     41  CA  THR     7       6.963  19.390  11.753
1.00 15.99
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ATOM     42  CB  THR     7       6.093  20.345  12.625
1.00 16.25
ATOM     43  OG1 THR     7       5.671  19.625  13.799
1.00 16.71
ATOM     44  CG2 THR     7       4.986  20.941  11.844
1.00 16.56
ATOM     45  C   THR     7       6.671  19.383  10.249
1.00 17.13
ATOM     46  O   THR     7       7.171  20.278   9.526
1.00 16.03
ATOM     47  N   GLY     8       5.941  18.369   9.742
1.00 18.40
ATOM     48  CA  GLY     8       5.511  18.374   8.345
1.00 19.41
ATOM     49  C   GLY     8       4.948  17.020   7.930
1.00 20.80
ATOM     50  O   GLY     8       4.664  16.135   8.751
1.00 20.52
ATOM     51  N   TYR     9       4.868  16.856   6.621
1.00 23.10
ATOM     52  CA  TYR     9       4.319  15.669   5.985
1.00 24.71
ATOM     53  CB  TYR     9       3.106  15.980   5.152
1.00 23.68
ATOM     54  CG  TYR     9       1.919  16.159   6.053
1.00 23.21
ATOM     55  CD1 TYR     9       1.098  15.072   6.302
1.00 24.08
ATOM     56  CE1 TYR     9       0.066  15.221   7.201
1.00 24.07
ATOM     57  CD2 TYR     9       1.719  17.380   6.663
1.00 23.62
ATOM     58  CE2 TYR     9       0.686  17.525   7.566
1.00 24.15
ATOM     59  CZ  TYR     9      -0.118  16.437   7.827
1.00 23.90
ATOM     60  OH  TYR     9      -1.099  16.569   8.790
1.00 24.93
ATOM     61  C   TYR     9       5.307  15.026   5.047
1.00 26.87
ATOM     62  O   TYR     9       6.094  15.819   4.503
1.00 26.77
ATOM     63  N   SER    10       5.354  13.666   4.903
1.00 29.24
ATOM     64  CA  SER    10       6.074  13.019   3.786
1.00 31.30
ATOM     65  CB  SER    10       7.578  12.994   4.132
1.00 33.29
ATOM     66  OG  SER    10       8.099  14.175   3.415
1.00 36.05
ATOM     67  C   SER    10       5.687  11.658   3.167
1.00 31.30
ATOM     68  O   SER    10       6.443  10.761   2.742
1.00 32.28
ATOM     69  N   ASN    11       4.369  11.565   3.067
1.00 31.13
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ATOM      70  CA  ASN  11       3.633  10.727   2.104
1.00 29.54
ATOM      71  CB  ASN  11       4.369   9.574   1.411
1.00 31.69
ATOM      72  CG  ASN  11       3.862   9.653  -0.044
1.00 34.14
ATOM      73  OD1 ASN  11       2.642   9.754  -0.339
1.00 33.69
ATOM      74  ND2 ASN  11       4.801   9.795  -0.993
1.00 34.10
ATOM      75  C   ASN  11       2.473  10.070   2.756
1.00 27.90
ATOM      76  O   ASN  11       2.349   8.838   2.987
1.00 28.07
ATOM      77  N   GLY  12       1.757  11.137   3.151
1.00 24.30
ATOM      78  CA  GLY  12       0.547  10.965   3.909
1.00 22.09
ATOM      79  C   GLY  12       0.839  10.928   5.400
1.00 19.23
ATOM      80  O   GLY  12      -0.117  10.901   6.189
1.00 19.87
ATOM      81  N   TYR  13       2.112  10.925   5.828
1.00 16.68
ATOM      82  CA  TYR  13       2.364  10.901   7.256
1.00 13.96
ATOM      83  CB  TYR  13       3.311   9.730   7.672
1.00 14.86
ATOM      84  CG  TYR  13       2.641   8.380   7.359
1.00 16.63
ATOM      85  CD1 TYR  13       1.913   7.701   8.331
1.00 18.73
ATOM      86  CE1 TYR  13       1.274   6.499   7.999
1.00 19.94
ATOM      87  CD2 TYR  13       2.730   7.866   6.084
1.00 16.53
ATOM      88  CE2 TYR  13       2.101   6.680   5.740
1.00 17.13
ATOM      89  CZ  TYR  13       1.384   6.020   6.710
1.00 18.35
ATOM      90  OH  TYR  13       0.758   4.855   6.377
1.00 19.27
ATOM      91  C   TYR  13       2.988  12.206   7.686
1.00 12.85
ATOM      92  O   TYR  13       3.768  12.869   7.013
1.00 11.47
ATOM      93  N   TYR  14       2.469  12.560   8.828
1.00 14.13
ATOM      94  CA  TYR  14       2.901  13.680   9.583
1.00 12.57
ATOM      95  CB  TYR  14       1.785  13.965  10.526
1.00 13.51
ATOM      96  CG  TYR  14       2.119  15.111  11.446
1.00 15.53
ATOM      97  CD1 TYR  14       2.055  16.416  10.949
1.00 16.07
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ATOM	98	CE1	TYR	14	2.374	17.464	11.785	
1.00	15.89							
ATOM	99	CD2	TYR	14	2.497	14.849	12.762	
1.00	16.13							
ATOM	100	CE2	TYR	14	2.827	15.898	13.594	
1.00	16.08							
ATOM	101	CZ	TYR	14	2.748	17.186	13.077	
1.00	16.73							
ATOM	102	OH	TYR	14	3.055	18.266	13.866	
1.00	18.42							
ATOM	103	C	TYR	14	4.165	13.171	10.274	
1.00	13.04							
ATOM	104	O	TYR	14	4.290	11.986	10.656	
1.00	11.98							
ATOM	105	N	TYR	15	5.107	14.068	10.452	
1.00	11.94							
ATOM	106	CA	TYR	15	6.302	13.775	11.193	
1.00	12.73							
ATOM	107	CB	TYR	15	7.551	13.590	10.265	
1.00	15.06							
ATOM	108	CG	TYR	15	8.113	14.812	9.520	
1.00	17.13							
ATOM	109	CD1	TYR	15	7.729	15.025	8.221	
1.00	17.27							
ATOM	110	CE1	TYR	15	8.165	16.146	7.558	
1.00	18.57							
ATOM	111	CD2	TYR	15	8.954	15.714	10.164	
1.00	18.44							
ATOM	112	CE2	TYR	15	9.389	16.845	9.506	
1.00	18.53							
ATOM	113	CZ	TYR	15	8.983	17.056	8.207	
1.00	19.40							
ATOM	114	OH	TYR	15	9.324	18.241	7.554	
1.00	21.86							
ATOM	115	C	TYR	15	6.470	14.984	12.096	
1.00	12.96							
ATOM	116	O	TYR	15	6.048	16.114	11.732	
1.00	10.33							
ATOM	117	N	SER	16	7.095	14.681	13.245	
1.00	11.29							
ATOM	118	CA	SER	16	7.450	15.700	14.199	
1.00	11.41							
ATOM	119	CB	SER	16	6.416	15.772	15.316	
1.00	9.89							
ATOM	120	OG	SER	16	6.764	16.764	16.281	
1.00	10.30							
ATOM	121	C	SER	16	8.790	15.268	14.773	
1.00	12.00							
ATOM	122	O	SER	16	8.944	14.112	15.217	
1.00	13.12							
ATOM	123	N	TYR	17	9.787	16.126	14.795	
1.00	10.82							
ATOM	124	CA	TYR	17	11.047	15.823	15.405	
1.00	8.67							
ATOM	125	CB	TYR	17	12.079	15.633	14.307	
1.00	7.15							

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ATOM    126  CG  TYR  17      13.533  15.647  14.771
1.00  7.03
ATOM    127  CD1 TYR  17      13.886  15.191  16.039
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ATOM    128  CE1 TYR  17      15.186  15.210  16.456
1.00  7.12
ATOM    129  CD2 TYR  17      14.502  16.123  13.920
1.00  6.69
ATOM    130  CE2 TYR  17      15.808  16.140  14.325
1.00  7.74
ATOM    131  CZ  TYR  17      16.131  15.685  15.596
1.00  7.93
ATOM    132  OH  TYR  17      17.422  15.747  16.038
1.00  7.83
ATOM    133  C   TYR  17      11.329  17.058  16.262
1.00 10.23
ATOM    134  O   TYR  17      11.442  18.156  15.702
1.00 11.78
ATOM    135  N   TRP  18      11.511  16.898  17.564
1.00 11.36
ATOM    136  CA  TRP  18      11.782  17.983  18.516
1.00 13.34
ATOM    137  CB  TRP  18      10.624  18.138  19.571
1.00 13.58
ATOM    138  CG  TRP  18      11.022  19.043  20.770
1.00 16.90
ATOM    139  CD2 TRP  18      11.668  18.676  21.973
1.00 15.25
ATOM    140  CE2 TRP  18      11.923  19.930  22.560
1.00 14.76
ATOM    141  CE3 TRP  18      12.079  17.518  22.620
1.00 14.38
ATOM    142  CD1 TRP  18      10.884  20.432  20.676
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ATOM    143  NE1 TRP  18      11.444  20.923  21.780
1.00 15.81
ATOM    144  CZ2 TRP  18      12.581  20.047  23.752
1.00 12.46
ATOM    145  CZ3 TRP  18      12.739  17.626  23.821
1.00 14.47
ATOM    146  CH2 TRP  18      12.988  18.880  24.379
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ATOM    147  C   TRP  18      13.076  17.609  19.261
1.00 14.41
ATOM    148  O   TRP  18      13.287  16.436  19.642
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ATOM    149  N   ASN  19      13.984  18.545  19.526
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ATOM    150  CA  ASN  19      15.035  18.250  20.451
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ATOM    151  CB  ASN  19      16.224  17.600  19.699
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ATOM    152  CG  ASN  19      17.151  18.440  18.894
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ATOM    153  OD1 ASN  19      17.244  19.629  19.124
1.00 18.68
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ATOM    154  ND2 ASN  19      17.864  17.941  17.903
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ATOM    155  C   ASN  19      15.396  19.550  21.166
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ATOM    156  O   ASN  19      14.989  20.636  20.741
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ATOM    157  N   ASP  20      16.131  19.441  22.251
1.00 11.88
ATOM    158  CA  ASP  20      16.504  20.540  23.135
1.00 11.46
ATOM    159  CB  ASP  20      16.797  19.893  24.518
1.00 10.44
ATOM    160  CG  ASP  20      18.025  18.968  24.621
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ATOM    161  OD1 ASP  20      18.718  18.720  23.650
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ATOM    162  OD2 ASP  20      18.310  18.428  25.678
1.00 10.16
ATOM    163  C   ASP  20      17.695  21.403  22.637
1.00 11.25
ATOM    164  O   ASP  20      18.103  22.388  23.291
1.00 13.63
ATOM    165  N   GLY  21      18.234  21.045  21.466
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ATOM    166  CA  GLY  21      19.380  21.652  20.875
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ATOM    167  C   GLY  21      20.648  20.856  21.103
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ATOM    168  O   GLY  21      21.648  21.119  20.438
1.00  8.94
ATOM    169  N   HIS  22      20.741  19.936  22.053
1.00  9.49
ATOM    170  CA  HIS  22      21.960  19.155  22.258
1.00  8.32
ATOM    171  CB  HIS  22      21.741  18.177  23.358
1.00  8.11
ATOM    172  CG  HIS  22      22.979  17.601  23.957
1.00  6.81
ATOM    173  CD2 HIS  22      23.428  18.016  25.187
1.00  6.67
ATOM    174  ND1 HIS  22      23.803  16.685  23.489
1.00  8.56
ATOM    175  CE1 HIS  22      24.747  16.526  24.386
1.00  8.59
ATOM    176  NE2 HIS  22      24.505  17.334  25.395
1.00  8.33
ATOM    177  C   HIS  22      22.310  18.377  20.984
1.00  8.28
ATOM    178  O   HIS  22      21.423  17.986  20.218
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ATOM    179  N   ALA  23      23.598  18.178  20.750
1.00  6.80
ATOM    180  CA  ALA  23      24.077  17.456  19.601
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ATOM    181  CB  ALA  23      25.543  17.722  19.414
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ATOM    182  C   ALA  23      23.884  15.951  19.853
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ATOM    183  O   ALA  23      23.555  15.546  20.983
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ATOM    184  N   GLY  24      24.053  15.132  18.813
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ATOM    185  CA  GLY  24      24.056  13.696  19.026
1.00 11.28
ATOM    186  C   GLY  24      23.003  12.932  18.274
1.00 12.70
ATOM    187  O   GLY  24      23.144  11.705  18.119
1.00 14.05
ATOM    188  N   VAL  25      21.954  13.601  17.782
1.00 11.17
ATOM    189  CA  VAL  25      20.870  12.933  17.087
1.00 11.42
ATOM    190  CB  VAL  25      19.513  13.475  17.632
1.00 10.39
ATOM    191  CG1 VAL  25      18.430  12.519  17.220
1.00  9.36
ATOM    192  CG2 VAL  25      19.568  13.692  19.147
1.00 10.78
ATOM    193  C   VAL  25      20.893  13.113  15.566
1.00 11.41
ATOM    194  O   VAL  25      21.215  14.214  15.055
1.00 10.88
ATOM    195  N   THR  26      20.545  12.071  14.797
1.00 12.50
ATOM    196  CA  THR  26      20.256  12.290  13.397
1.00 12.14
ATOM    197  CB  THR  26      21.498  11.864  12.462
1.00 13.95
ATOM    198  OG1 THR  26      21.030  10.946  11.485
1.00 16.06
ATOM    199  CG2 THR  26      22.706  11.411  13.230
1.00 12.20
ATOM    200  C   THR  26      18.959  11.552  13.126
1.00 12.76
ATOM    201  O   THR  26      18.758  10.385  13.519
1.00 13.16
ATOM    202  N   TYR  27      17.998  12.359  12.665
1.00 10.77
ATOM    203  CA  TYR  27      16.665  11.919  12.311
1.00 10.37
ATOM    204  CB  TYR  27      15.581  12.842  12.861
1.00  9.29
ATOM    205  CG  TYR  27      14.148  12.477  12.477
1.00  8.75
ATOM    206  CD1 TYR  27      13.599  12.815  11.240
1.00  9.08
ATOM    207  CE1 TYR  27      12.306  12.458  10.929
1.00 10.58
ATOM    208  CD2 TYR  27      13.378  11.789  13.377
1.00  8.03
ATOM    209  CE2 TYR  27      12.077  11.427  13.078
1.00  8.62
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ATOM    210  CZ  TYR  27      11.547  11.760  11.854  1.00 10.33
ATOM    211  OH  TYR  27      10.269  11.337  11.544  1.00 11.41
ATOM    212  C   TYR  27      16.526  11.902  10.789  1.00 10.52
ATOM    213  O   TYR  27      16.581  12.905  10.043  1.00  9.97
ATOM    214  N   THR  28      16.226  10.719  10.289  1.00 10.67
ATOM    215  CA  THR  28      15.990  10.624   8.866  1.00 13.02
ATOM    216  CB  THR  28      17.060   9.679   8.276  1.00 12.93
ATOM    217  OG1 THR  28      18.323  10.097   8.775  1.00 14.75
ATOM    218  CG2 THR  28      17.051   9.700   6.740  1.00 12.92
ATOM    219  C   THR  28      14.577  10.154   8.537  1.00 13.28
ATOM    220  O   THR  28      14.095   9.159   9.058  1.00 11.76
ATOM    221  N   ASN  29      13.877  10.924   7.712  1.00 15.11
ATOM    222  CA  ASN  29      12.610  10.493   7.127  1.00 17.11
ATOM    223  CB  ASN  29      11.759  11.680   6.583  1.00 20.17
ATOM    224  CG  ASN  29      10.957  11.330   5.310  1.00 23.95
ATOM    225  OD1 ASN  29       9.752  11.006   5.349  1.00 25.19
ATOM    226  ND2 ASN  29      11.595  11.263   4.123  1.00 26.04
ATOM    227  C   ASN  29      12.949   9.585   5.931  1.00 17.45
ATOM    228  O   ASN  29      13.733   9.976   5.034  1.00 13.40
ATOM    229  N   GLY  30      12.292   8.430   5.973  1.00 17.19
ATOM    230  CA  GLY  30      12.366   7.507   4.876  1.00 20.59
ATOM    231  C   GLY  30      11.061   7.603   4.091  1.00 20.60
ATOM    232  O   GLY  30      10.149   8.393   4.394  1.00 21.82
ATOM    233  N   GLY  31      10.880   6.783   3.065  1.00 20.69
ATOM    234  CA  GLY  31       9.593   6.864   2.353  1.00 21.04
ATOM    235  C   GLY  31       8.379   6.433   3.198  1.00 20.17
ATOM    236  O   GLY  31       8.534   5.626   4.140  1.00 21.45
ATOM    237  N   GLY  32       7.192   7.004   2.901  1.00 17.60
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ATOM    238  CA  GLY  32      5.921   6.633   3.522
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ATOM    239  C   GLY  32      5.950   6.688   5.053
1.00 13.27
ATOM    240  O   GLY  32      6.228   7.744   5.678
1.00 14.99
ATOM    241  N   GLY  33      5.743   5.536   5.682
1.00 10.93
ATOM    242  CA  GLY  33      5.684   5.504   7.127
1.00  9.50
ATOM    243  C   GLY  33      7.020   5.351   7.849
1.00  7.59
ATOM    244  O   GLY  33      7.024   5.171   9.070
1.00  6.40
ATOM    245  N   SER  34      8.144   5.399   7.165
1.00  7.69
ATOM    246  CA  SER  34      9.433   5.095   7.768
1.00  9.77
ATOM    247  CB  SER  34     10.348   4.331   6.832
1.00  9.92
ATOM    248  OG  SER  34      9.840   3.098   6.383
1.00 11.79
ATOM    249  C   SER  34     10.278   6.253   8.213
1.00  8.85
ATOM    250  O   SER  34     10.312   7.296   7.519
1.00 11.44
ATOM    251  N   PHE  35     10.938   6.029   9.340
1.00  8.14
ATOM    252  CA  PHE  35     11.958   6.938   9.798
1.00  7.22
ATOM    253  CB  PHE  35     11.353   8.045  10.667
1.00  5.37
ATOM    254  CG  PHE  35     10.780   7.622  11.990
1.00  4.40
ATOM    255  CD1 PHE  35      9.482   7.189  12.055
1.00  5.29
ATOM    256  CD2 PHE  35     11.578   7.664  13.111
1.00  2.70
ATOM    257  CE1 PHE  35      8.972   6.789  13.273
1.00  5.43
ATOM    258  CE2 PHE  35     11.073   7.267  14.314
1.00  4.23
ATOM    259  CZ  PHE  35      9.772   6.828  14.399
1.00  5.42
ATOM    260  C   PHE  35     12.974   6.128  10.596
1.00  7.01
ATOM    261  O   PHE  35     12.624   5.078  11.148
1.00  7.90
ATOM    262  N   THR  36     14.193   6.657  10.651
1.00  6.93
ATOM    263  CA  THR  36     15.346   6.181  11.379
1.00  8.26
ATOM    264  CB  THR  36     16.530   5.882  10.407
1.00  9.23
ATOM    265  OG1 THR  36     15.996   5.076   9.379
1.00 10.78
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ATOM    266  CG2 THR  36      17.701   5.125  11.030
1.00 10.22
ATOM    267  C   THR  36      15.758   7.312  12.343
1.00  9.84
ATOM    268  O   THR  36      15.537   8.507  12.050
1.00  9.61
ATOM    269  N   VAL  37      16.298   6.977  13.520
1.00  9.80
ATOM    270  CA  VAL  37      16.826   7.961  14.439
1.00  9.70
ATOM    271  CB  VAL  37      15.976   8.312  15.692
1.00 10.77
ATOM    272  CG1 VAL  37      16.624   9.555  16.323
1.00 10.77
ATOM    273  CG2 VAL  37      14.509   8.571  15.389
1.00 10.75
ATOM    274  C   VAL  37      18.010   7.180  14.958
1.00 10.19
ATOM    275  O   VAL  37      17.903   5.987  15.292
1.00 12.44
ATOM    276  N   ASN  38      19.154   7.842  15.003
1.00  9.51
ATOM    277  CA  ASN  38      20.363   7.273  15.517
1.00 10.83
ATOM    278  CB  ASN  38      21.351   7.007  14.392
1.00 13.08
ATOM    279  CG  ASN  38      20.884   5.745  13.683
1.00 18.12
ATOM    280  OD1 ASN  38      20.079   5.762  12.722
1.00 18.42
ATOM    281  ND2 ASN  38      21.365   4.580  14.159
1.00 18.73
ATOM    282  C   ASN  38      20.878   8.342  16.455
1.00 10.89
ATOM    283  O   ASN  38      20.817   9.510  16.083
1.00 10.36
ATOM    284  N   TRP  39      21.339   8.018  17.651
1.00  8.76
ATOM    285  CA  TRP  39      21.772   9.048  18.577
1.00 10.30
ATOM    286  CB  TRP  39      20.583   9.478  19.449
1.00  8.23
ATOM    287  CG  TRP  39      19.943   8.426  20.359
1.00  7.73
ATOM    288  CD2 TRP  39      19.034   7.452  19.994
1.00  7.65
ATOM    289  CE2 TRP  39      18.728   6.903  21.234
1.00  7.27
ATOM    290  CE3 TRP  39      18.410   6.956  18.835
1.00  8.18
ATOM    291  CD1 TRP  39      20.183   8.466  21.706
1.00  5.84
ATOM    292  NE1 TRP  39      19.425   7.538  22.200
1.00  5.45
ATOM    293  CZ2 TRP  39      17.805   5.847  21.320
1.00  6.19
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ATOM    294  CZ3 TRP  39      17.487   5.910  18.934
1.00  6.98
ATOM    295  CH2 TRP  39      17.194   5.367  20.174
1.00  6.37
ATOM    296  C   TRP  39      22.898   8.504  19.411
1.00 10.96
ATOM    297  O   TRP  39      22.879   7.317  19.771
1.00 13.55
ATOM    298  N   SER  40      23.916   9.328  19.631
1.00 13.07
ATOM    299  CA  SER  40      25.061   9.010  20.463
1.00 13.74
ATOM    300  CB  SER  40      26.357   8.815  19.658
1.00 14.37
ATOM    301  OG  SER  40      26.111   7.978  18.518
1.00 20.42
ATOM    302  C   SER  40      25.227  10.244  21.326
1.00 12.72
ATOM    303  O   SER  40      25.357  11.332  20.770
1.00 10.52
ATOM    304  N   ASN  41      25.165  10.040  22.646
1.00 13.35
ATOM    305  CA  ASN  41      25.338  11.092  23.656
1.00 13.53
ATOM    306  CB  ASN  41      26.757  11.568  23.695
1.00 15.67
ATOM    307  CG  ASN  41      27.771  10.495  24.008
1.00 18.65
ATOM    308  OD1 ASN  41      28.757  10.358  23.282
1.00 22.59
ATOM    309  ND2 ASN  41      27.653   9.675  25.050
1.00 19.49
ATOM    310  C   ASN  41      24.441  12.287  23.350
1.00 12.66
ATOM    311  O   ASN  41      24.873  13.422  23.078
1.00 11.83
ATOM    312  N   SER  42      23.155  11.974  23.365
1.00 10.43
ATOM    313  CA  SER  42      22.156  12.925  22.935
1.00 10.63
ATOM    314  CB  SER  42      20.894  12.124  22.555
1.00  9.58
ATOM    315  OG  SER  42      20.288  11.609  23.751
1.00  8.86
ATOM    316  C   SER  42      21.856  13.905  24.087
1.00  9.67
ATOM    317  O   SER  42      22.501  13.874  25.146
1.00 11.35
ATOM    318  N   GLY  43      20.855  14.769  23.867
1.00  9.05
ATOM    319  CA  GLY  43      20.215  15.460  24.946
1.00  6.67
ATOM    320  C   GLY  43      18.840  14.815  25.018
1.00  7.83
ATOM    321  O   GLY  43      18.694  13.583  24.897
1.00  9.11
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ATOM    322  N   ASN  44      17.809  15.616  25.181
1.00  6.80
ATOM    323  CA  ASN  44      16.450  15.160  25.134
1.00  8.62
ATOM    324  CB  ASN  44      15.643  15.843  26.251
1.00  9.32
ATOM    325  CG  ASN  44      14.239  15.305  26.407
1.00 11.76
ATOM    326  OD1 ASN  44      13.395  16.034  26.942
1.00 14.29
ATOM    327  ND2 ASN  44      13.867  14.062  26.021
1.00 12.06
ATOM    328  C   ASN  44      15.906  15.528  23.737
1.00  8.95
ATOM    329  O   ASN  44      16.070  16.664  23.240
1.00  7.76
ATOM    330  N   PHE  45      15.358  14.499  23.068
1.00  8.42
ATOM    331  CA  PHE  45      14.723  14.592  21.745
1.00  8.92
ATOM    332  CB  PHE  45      15.707  14.187  20.598
1.00  6.83
ATOM    333  CG  PHE  45      16.041  12.710  20.589
1.00  7.80
ATOM    334  CD1 PHE  45      15.274  11.836  19.819
1.00  7.09
ATOM    335  CD2 PHE  45      17.051  12.228  21.403
1.00  7.62
ATOM    336  CE1 PHE  45      15.515  10.477  19.875
1.00  7.59
ATOM    337  CE2 PHE  45      17.274  10.857  21.433
1.00  7.17
ATOM    338  CZ  PHE  45      16.510   9.994  20.678
1.00  5.89
ATOM    339  C   PHE  45      13.491  13.660  21.684
1.00  8.27
ATOM    340  O   PHE  45      13.435  12.692  22.478
1.00  9.67
ATOM    341  N   VAL  46      12.541  13.912  20.755
1.00  7.23
ATOM    342  CA  VAL  46      11.346  13.130  20.533
1.00  7.92
ATOM    343  CB  VAL  46      10.076  13.715  21.179
1.00  8.74
ATOM    344  CG1 VAL  46       8.860  12.823  20.921
1.00  9.06
ATOM    345  CG2 VAL  46      10.226  13.721  22.656
1.00  8.48
ATOM    346  C   VAL  46      11.179  13.174  19.034
1.00  9.06
ATOM    347  O   VAL  46      11.069  14.250  18.422
1.00  7.02
ATOM    348  N   ALA  47      11.148  11.984  18.423
1.00 10.19
ATOM    349  CA  ALA  47      11.088  11.923  16.970
1.00 11.40
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ATOM    350  CB  ALA  47      12.487  11.628  16.548
1.00 14.35
ATOM    351  C   ALA  47      10.115  10.894  16.379
1.00 11.87
ATOM    352  O   ALA  47      10.126   9.770  16.905
1.00 11.08
ATOM    353  N   GLY  48       9.315  11.134  15.349
1.00  9.25
ATOM    354  CA  GLY  48       8.396  10.089  14.949
1.00 11.87
ATOM    355  C   GLY  48       7.493  10.425  13.794
1.00 11.00
ATOM    356  O   GLY  48       7.513  11.559  13.316
1.00 11.18
ATOM    357  N   LYS  49       6.712   9.448  13.324
1.00  9.47
ATOM    358  CA  LYS  49       5.783   9.700  12.233
1.00  8.49
ATOM    359  CB  LYS  49       6.156   8.995  10.959
1.00  7.23
ATOM    360  CG  LYS  49       7.203   9.663  10.124
1.00  7.71
ATOM    361  CD  LYS  49       7.411   8.921   8.809
1.00  8.83
ATOM    362  CE  LYS  49       8.141   9.814   7.800
1.00  9.61
ATOM    363  NZ  LYS  49       8.376   9.080   6.554
1.00 11.96
ATOM    364  C   LYS  49       4.398   9.237  12.597
1.00  8.57
ATOM    365  O   LYS  49       4.248   8.384  13.486
1.00  9.28
ATOM    366  N   GLY  50       3.361   9.766  11.960
1.00  8.33
ATOM    367  CA  GLY  50       2.000   9.425  12.301
1.00  8.35
ATOM    368  C   GLY  50       0.963  10.342  11.663
1.00 10.55
ATOM    369  O   GLY  50       0.966  10.545  10.442
1.00 10.22
ATOM    370  N   TRP  51       0.070  10.906  12.511
1.00 11.01
ATOM    371  CA  TRP  51      -1.118  11.652  12.085
1.00 12.06
ATOM    372  CB  TRP  51      -2.423  10.864  12.333
1.00 11.79
ATOM    373  CG  TRP  51      -2.333   9.504  11.635
1.00 13.30
ATOM    374  CD2 TRP  51      -1.674   8.392  12.124
1.00 13.13
ATOM    375  CE2 TRP  51      -1.810   7.489  11.080
1.00 14.19
ATOM    376  CE3 TRP  51      -0.996   8.040  13.282
1.00 12.35
ATOM    377  CD1 TRP  51      -2.848   9.301  10.393
1.00 13.99
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ATOM    378  NE1 TRP  51      -2.494   8.051  10.078
1.00 14.34
ATOM    379  CZ2 TRP  51      -1.271   6.217  11.162
1.00 15.05
ATOM    380  CZ3 TRP  51      -0.457   6.769  13.365
1.00 14.41
ATOM    381  CH2 TRP  51      -0.595   5.873  12.316
1.00 14.24
ATOM    382  C   TRP  51      -1.291  12.983  12.788
1.00 11.13
ATOM    383  O   TRP  51      -0.968  13.135  13.964
1.00  9.97
ATOM    384  N   GLN  52      -1.795  13.962  12.028
1.00 13.14
ATOM    385  CA  GLN  52      -2.174  15.267  12.560
1.00 14.01
ATOM    386  CB  GLN  52      -1.088  16.284  12.168
1.00 13.62
ATOM    387  CG  GLN  52      -1.328  17.699  12.606
1.00 15.07
ATOM    388  CD  GLN  52      -1.494  17.915  14.104
1.00 14.27
ATOM    389  OE1 GLN  52      -0.629  18.407  14.830
1.00 16.02
ATOM    390  NE2 GLN  52      -2.657  17.626  14.635
1.00 13.08
ATOM    391  C   GLN  52      -3.559  15.701  12.054
1.00 13.05
ATOM    392  O   GLN  52      -3.704  16.030  10.868
1.00 14.72
ATOM    393  N   PRO  53      -4.628  15.721  12.866
1.00 13.11
ATOM    394  CD  PRO  53      -5.967  16.150  12.469
1.00 13.04
ATOM    395  CA  PRO  53      -4.680  15.205  14.223
1.00 12.06
ATOM    396  CB  PRO  53      -5.876  15.851  14.828
1.00 12.56
ATOM    397  CG  PRO  53      -6.846  15.722  13.670
1.00 12.67
ATOM    398  C   PRO  53      -4.799  13.691  14.254
1.00 12.14
ATOM    399  O   PRO  53      -5.135  13.011  13.248
1.00 10.91
ATOM    400  N   GLY  54      -4.607  13.246  15.484
1.00 12.13
ATOM    401  CA  GLY  54      -4.869  11.866  15.739
1.00 13.75
ATOM    402  C   GLY  54      -6.346  11.663  16.041
1.00 15.01
ATOM    403  O   GLY  54      -7.172  12.582  16.005
1.00 15.77
ATOM    404  N   THR  55      -6.740  10.453  16.400
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ATOM    405  CA  THR  55      -8.135  10.178  16.694
1.00 16.04
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ATOM 406 CB THR 55 -8.827 9.410 15.572 1.00 15.48
ATOM 407 OG1 THR 55 -8.200 8.132 15.474 1.00 15.49
ATOM 408 CG2 THR 55 -8.800 10.183 14.265 1.00 15.96
ATOM 409 C THR 55 -8.258 9.350 17.937 1.00 16.77
ATOM 410 O THR 55 -7.322 8.644 18.310 1.00 16.68
ATOM 411 N LYS 56 -9.438 9.279 18.531 1.00 19.29
ATOM 412 CA LYS 56 -9.564 8.452 19.725 1.00 21.92
ATOM 413 CB LYS 56 -10.675 9.096 20.595 1.00 23.92
ATOM 414 CG LYS 56 -10.234 10.565 20.893 1.00 26.95
ATOM 415 CD LYS 56 -10.828 11.213 22.151 1.00 29.47
ATOM 416 CE LYS 56 -10.528 12.739 22.187 1.00 31.18
ATOM 417 NZ LYS 56 -10.775 13.356 23.492 1.00 32.24
ATOM 418 C LYS 56 -9.831 6.988 19.365 1.00 22.96
ATOM 419 O LYS 56 -10.122 6.137 20.230 1.00 24.28
ATOM 420 N ASN 57 -9.656 6.581 18.102 1.00 22.55
ATOM 421 CA ASN 57 -10.009 5.214 17.735 1.00 22.75
ATOM 422 CB ASN 57 -11.479 5.164 17.184 1.00 24.35
ATOM 423 CG ASN 57 -11.671 5.923 15.848 1.00 27.06
ATOM 424 OD1 ASN 57 -11.050 6.960 15.544 1.00 27.62
ATOM 425 ND2 ASN 57 -12.539 5.461 14.955 1.00 27.78
ATOM 426 C ASN 57 -9.048 4.680 16.679 1.00 21.44
ATOM 427 O ASN 57 -9.397 3.777 15.896 1.00 22.71
ATOM 428 N LYS 58 -7.821 5.193 16.624 1.00 18.17
ATOM 429 CA LYS 58 -6.950 4.785 15.552 1.00 15.67
ATOM 430 CB LYS 58 -5.911 5.860 15.363 1.00 16.07
ATOM 431 CG LYS 58 -5.278 5.787 13.998 1.00 17.83
ATOM 432 CD LYS 58 -4.526 7.069 13.607 1.00 18.91
ATOM 433 CE LYS 58 -5.410 8.299 13.205 1.00 18.54

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ATOM    434  NZ  LYS    58      -6.249   8.140  11.995
1.00 19.60
ATOM    435  C   LYS    58      -6.334   3.476  15.978
1.00 13.93
ATOM    436  O   LYS    58      -6.086   3.300  17.171
1.00 15.02
ATOM    437  N   VAL    59      -6.174   2.474  15.131
1.00 13.40
ATOM    438  CA  VAL    59      -5.364   1.313  15.506
1.00 12.92
ATOM    439  CB  VAL    59      -6.203  -0.053  15.388
1.00 14.48
ATOM    440  CG1 VAL    59      -7.120  -0.117  14.200
1.00 16.35
ATOM    441  CG2 VAL    59      -5.192  -1.198  15.311
1.00 13.23
ATOM    442  C   VAL    59      -4.138   1.408  14.583
1.00 11.86
ATOM    443  O   VAL    59      -4.167   1.542  13.343
1.00 12.38
ATOM    444  N   ILE    60      -3.061   1.577  15.346
1.00 10.27
ATOM    445  CA  ILE    60      -1.749   1.915  14.826
1.00  9.01
ATOM    446  CB  ILE    60      -1.119   3.011  15.700
1.00  8.58
ATOM    447  CG2 ILE    60       0.280   3.326  15.213
1.00  8.04
ATOM    448  CG1 ILE    60      -2.000   4.252  15.663
1.00  7.60
ATOM    449  CD1 ILE    60      -1.611   5.192  16.759
1.00  9.90
ATOM    450  C   ILE    60      -0.851   0.725  14.800
1.00  9.88
ATOM    451  O   ILE    60      -0.680   0.059  15.822
1.00  9.11
ATOM    452  N   ASN    61      -0.270   0.546  13.616
1.00 11.11
ATOM    453  CA  ASN    61       0.671  -0.545  13.341
1.00 10.36
ATOM    454  CB  ASN    61       0.366  -1.204  12.006
1.00 12.63
ATOM    455  CG  ASN    61      -1.072  -1.619  11.996
1.00 14.76
ATOM    456  OD1 ASN    61      -1.478  -2.378  12.874
1.00 16.78
ATOM    457  ND2 ASN    61      -1.921  -1.111  11.095
1.00 15.45
ATOM    458  C   ASN    61       2.088  -0.032  13.291
1.00  9.66
ATOM    459  O   ASN    61       2.338   1.095  12.867
1.00  9.13
ATOM    460  N   PHE    62       3.042  -0.806  13.765
1.00  9.56
ATOM    461  CA  PHE    62       4.422  -0.405  13.688
1.00 10.44
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ATOM    462  CB  PHE  62      4.865   0.454  14.933
1.00  9.48
ATOM    463  CG  PHE  62      4.657  -0.145  16.314
1.00  9.74
ATOM    464  CD1 PHE  62      5.751  -0.618  17.021
1.00 10.19
ATOM    465  CD2 PHE  62      3.371  -0.259  16.828
1.00 10.24
ATOM    466  CE1 PHE  62      5.546  -1.224  18.250
1.00  9.20
ATOM    467  CE2 PHE  62      3.176  -0.867  18.054
1.00  9.66
ATOM    468  CZ  PHE  62      4.274  -1.347  18.757
1.00  9.91
ATOM    469  C   PHE  62      5.348  -1.590  13.570
1.00  9.69
ATOM    470  O   PHE  62      5.066  -2.608  14.200
1.00 11.84
ATOM    471  N   SER  63      6.404  -1.538  12.767
1.00 11.03
ATOM    472  CA  SER  63      7.449  -2.547  12.847
1.00 11.59
ATOM    473  CB  SER  63      7.328  -3.650  11.765
1.00 12.00
ATOM    474  OG  SER  63      7.114  -3.195  10.463
1.00 12.51
ATOM    475  C   SER  63      8.819  -1.923  12.715
1.00 11.22
ATOM    476  O   SER  63      8.928  -0.727  12.379
1.00 12.00
ATOM    477  N   GLY  64      9.823  -2.781  12.942
1.00 10.23
ATOM    478  CA  GLY  64     11.193  -2.372  12.823
1.00  8.33
ATOM    479  C   GLY  64     11.988  -2.761  14.029
1.00  7.41
ATOM    480  O   GLY  64     11.615  -3.676  14.751
1.00  8.00
ATOM    481  N   SER  65     13.077  -2.058  14.222
1.00  7.92
ATOM    482  CA  SER  65     13.961  -2.346  15.314
1.00 10.77
ATOM    483  CB  SER  65     15.240  -2.865  14.680
1.00 12.34
ATOM    484  OG  SER  65     15.702  -2.010  13.636
1.00 12.01
ATOM    485  C   SER  65     14.215  -1.151  16.250
1.00 10.66
ATOM    486  O   SER  65     14.393  -0.010  15.791
1.00 11.27
ATOM    487  N   TYR  66     14.338  -1.407  17.535
1.00  8.76
ATOM    488  CA  TYR  66     14.573  -0.359  18.514
1.00  8.36
ATOM    489  CB  TYR  66     13.258  -0.091  19.279
1.00  6.60
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ATOM    490  CG  TYR  66      13.429   0.738  20.539
1.00  5.88
ATOM    491  CD1 TYR  66      13.005   0.198  21.732
1.00  3.88
ATOM    492  CE1 TYR  66      13.053   0.928  22.894
1.00  3.78
ATOM    493  CD2 TYR  66      13.932   2.032  20.506
1.00  5.67
ATOM    494  CE2 TYR  66      13.982   2.780  21.677
1.00  3.65
ATOM    495  CZ  TYR  66      13.533   2.217  22.851
1.00  4.38
ATOM    496  OH  TYR  66      13.501   2.961  24.006
1.00  5.96
ATOM    497  C   TYR  66      15.667  -0.905  19.416
1.00  8.49
ATOM    498  O   TYR  66      15.539  -1.954  20.045
1.00  8.59
ATOM    499  N   ASN  67      16.768  -0.197  19.470
1.00 10.09
ATOM    500  CA  ASN  67      17.951  -0.686  20.156
1.00 11.81
ATOM    501  CB  ASN  67      18.968  -1.158  19.130
1.00 15.94
ATOM    502  CG  ASN  67      18.427  -2.110  18.028
1.00 20.43
ATOM    503  OD1 ASN  67      18.079  -3.269  18.260
1.00 21.28
ATOM    504  ND2 ASN  67      18.287  -1.681  16.767
1.00 22.48
ATOM    505  C   ASN  67      18.556   0.438  21.016
1.00 11.16
ATOM    506  O   ASN  67      19.512   1.103  20.618
1.00  8.26
ATOM    507  N   PRO  68      17.937   0.733  22.173
1.00 10.94
ATOM    508  CD  PRO  68      16.694   0.131  22.659
1.00 10.82
ATOM    509  CA  PRO  68      18.378   1.755  23.099
1.00 11.97
ATOM    510  CB  PRO  68      17.103   2.066  23.890
1.00 11.53
ATOM    511  CG  PRO  68      16.437   0.732  24.019
1.00 10.89
ATOM    512  C   PRO  68      19.561   1.340  23.957
1.00 11.57
ATOM    513  O   PRO  68      19.689   0.209  24.442
1.00 10.78
ATOM    514  N   ASN  69      20.429   2.277  24.207
1.00  9.77
ATOM    515  CA  ASN  69      21.523   2.025  25.094
1.00 11.79
ATOM    516  CB  ASN  69      22.744   2.036  24.242
1.00 13.60
ATOM    517  CG  ASN  69      23.967   1.656  25.041
1.00 17.40
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ATOM    518  OD1 ASN    69      25.063   1.812  24.504
1.00 21.47
ATOM    519  ND2 ASN    69      24.006   1.089  26.244
1.00 17.05
ATOM    520  C   ASN    69      21.484   3.117  26.179
1.00 12.60
ATOM    521  O   ASN    69      22.208   4.144  26.160
1.00 13.38
ATOM    522  N   GLY    70      20.491   2.932  27.046
1.00  8.91
ATOM    523  CA  GLY    70      20.226   3.850  28.114
1.00  8.37
ATOM    524  C   GLY    70      18.795   4.357  28.073
1.00  8.21
ATOM    525  O   GLY    70      17.842   3.663  27.670
1.00  9.06
ATOM    526  N   ASN    71      18.656   5.595  28.502
1.00  7.03
ATOM    527  CA  ASN    71      17.347   6.161  28.701
1.00  7.69
ATOM    528  CB  ASN    71      17.448   7.409  29.589
1.00  7.29
ATOM    529  CG  ASN    71      16.104   7.886  30.142
1.00  8.85
ATOM    530  OD1 ASN    71      15.032   7.684  29.577
1.00  9.56
ATOM    531  ND2 ASN    71      16.047   8.491  31.329
1.00  7.50
ATOM    532  C   ASN    71      16.668   6.525  27.391
1.00  8.37
ATOM    533  O   ASN    71      17.072   7.537  26.762
1.00  9.29
ATOM    534  N   SER    72      15.643   5.733  27.043
1.00  6.66
ATOM    535  CA  SER    72      14.864   5.953  25.837
1.00  7.05
ATOM    536  CB  SER    72      15.699   5.398  24.662
1.00  7.68
ATOM    537  OG  SER    72      15.191   5.543  23.349
1.00  8.77
ATOM    538  C   SER    72      13.483   5.286  25.932
1.00  8.50
ATOM    539  O   SER    72      13.292   4.335  26.722
1.00  8.19
ATOM    540  N   TYR    73      12.520   5.701  25.110
1.00  7.79
ATOM    541  CA  TYR    73      11.190   5.154  25.058
1.00  7.74
ATOM    542  CB  TYR    73      10.094   6.073  25.659
1.00  7.66
ATOM    543  CG  TYR    73      10.319   6.583  27.099
1.00 10.11
ATOM    544  CD1 TYR    73      10.919   5.817  28.103
1.00  9.07
ATOM    545  CE1 TYR    73      11.121   6.358  29.366
1.00  9.86
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ATOM    546  CD2 TYR    73       9.920   7.881  27.376
1.00 10.46
ATOM    547  CE2 TYR    73      10.116   8.418  28.625
1.00  9.86
ATOM    548  CZ  TYR    73      10.709   7.668  29.607
1.00 10.88
ATOM    549  OH  TYR    73      10.868   8.246  30.846
1.00 10.66
ATOM    550  C   TYR    73      10.796   4.932  23.600
1.00  8.12
ATOM    551  O   TYR    73      11.244   5.627  22.676
1.00  8.31
ATOM    552  N   LEU    74       9.946   3.942  23.394
1.00  6.70
ATOM    553  CA  LEU    74       9.292   3.736  22.104
1.00  7.34
ATOM    554  CB  LEU    74       9.677   2.388  21.485
1.00  7.37
ATOM    555  CG  LEU    74       8.939   1.937  20.190
1.00  8.67
ATOM    556  CD1 LEU    74       9.182   2.886  19.023
1.00  8.01
ATOM    557  CD2 LEU    74       9.430   0.523  19.850
1.00  8.15
ATOM    558  C   LEU    74       7.805   3.752  22.500
1.00  6.80
ATOM    559  O   LEU    74       7.359   2.918  23.297
1.00  6.53
ATOM    560  N   SER    75       7.030   4.688  21.973
1.00  5.87
ATOM    561  CA  SER    75       5.669   4.839  22.425
1.00  5.53
ATOM    562  CB  SER    75       5.555   5.907  23.516
1.00  6.86
ATOM    563  OG  SER    75       6.582   5.955  24.476
1.00 11.37
ATOM    564  C   SER    75       4.770   5.295  21.297
1.00  4.71
ATOM    565  O   SER    75       5.235   5.772  20.263
1.00  4.19
ATOM    566  N   ILE    76       3.485   5.121  21.489
1.00  3.93
ATOM    567  CA  ILE    76       2.508   5.857  20.711
1.00  5.49
ATOM    568  CB  ILE    76       1.190   5.090  20.679
1.00  7.40
ATOM    569  CG2 ILE    76       0.068   6.005  20.340
1.00  8.03
ATOM    570  CG1 ILE    76       1.184   4.029  19.557
1.00  8.58
ATOM    571  CD1 ILE    76       2.241   2.921  19.633
1.00  9.45
ATOM    572  C   ILE    76       2.465   7.108  21.600
1.00  6.93
ATOM    573  O   ILE    76       2.370   6.973  22.829
1.00  7.49
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ATOM    574  N   TYR  77      2.627   8.295  21.020
1.00  6.57
ATOM    575  CA  TYR  77      2.811   9.555  21.724
1.00  5.67
ATOM    576  CB  TYR  77      4.299   9.948  21.647
1.00  4.54
ATOM    577  CG  TYR  77      4.686  11.364  22.139
1.00  4.19
ATOM    578  CD1 TYR  77      4.668  11.654  23.497
1.00  4.75
ATOM    579  CE1 TYR  77      4.991  12.901  23.966
1.00  4.71
ATOM    580  CD2 TYR  77      5.044  12.363  21.240
1.00  4.76
ATOM    581  CE2 TYR  77      5.373  13.638  21.694
1.00  3.95
ATOM    582  CZ  TYR  77      5.341  13.894  23.060
1.00  5.09
ATOM    583  OH  TYR  77      5.641  15.159  23.541
1.00  6.11
ATOM    584  C   TYR  77      1.961  10.639  21.098
1.00  5.81
ATOM    585  O   TYR  77      1.888  10.741  19.856
1.00  6.39
ATOM    586  N   GLY  78      1.359  11.509  21.898
1.00  5.96
ATOM    587  CA  GLY  78      0.615  12.588  21.278
1.00  6.69
ATOM    588  C   GLY  78      0.156  13.586  22.288
1.00  7.28
ATOM    589  O   GLY  78      0.537  13.494  23.466
1.00  7.84
ATOM    590  N   TRP  79     -0.672  14.528  21.822
1.00  7.76
ATOM    591  CA  TRP  79     -1.234  15.560  22.707
1.00  7.51
ATOM    592  CB  TRP  79     -0.598  16.949  22.555
1.00  6.56
ATOM    593  CG  TRP  79      0.876  17.041  22.832
1.00  5.50
ATOM    594  CD2 TRP  79      1.419  17.620  23.935
1.00  5.87
ATOM    595  CE2 TRP  79      2.774  17.469  23.695
1.00  5.84
ATOM    596  CE3 TRP  79      0.951  18.249  25.072
1.00  5.83
ATOM    597  CD1 TRP  79      1.819  16.560  21.962
1.00  6.72
ATOM    598  NE1 TRP  79      2.963  16.842  22.524
1.00  5.73
ATOM    599  CZ2 TRP  79      3.695  17.950  24.600
1.00  5.70
ATOM    600  CZ3 TRP  79      1.867  18.733  25.972
1.00  5.50
ATOM    601  CH2 TRP  79      3.226  18.581  25.742
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ATOM    602  C   TRP  79     -2.702  15.796  22.434
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ATOM    603  O   TRP  79     -3.219  15.434  21.365
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ATOM    604  N   SER  80     -3.360  16.381  23.440
1.00  8.56
ATOM    605  CA  SER  80     -4.683  16.933  23.242
1.00 11.11
ATOM    606  CB  SER  80     -5.761  16.138  23.974
1.00 10.43
ATOM    607  OG  SER  80     -5.890  16.355  25.385
1.00 16.05
ATOM    608  C   SER  80     -4.643  18.381  23.781
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ATOM    609  O   SER  80     -3.732  18.753  24.528
1.00  9.07
ATOM    610  N   ARG  81     -5.593  19.225  23.392
1.00 12.84
ATOM    611  CA  ARG  81     -5.732  20.562  23.953
1.00 15.11
ATOM    612  CB  ARG  81     -5.614  21.564  22.851
1.00 18.41
ATOM    613  CG  ARG  81     -4.205  21.639  22.307
1.00 22.70
ATOM    614  CD  ARG  81     -4.381  22.114  20.897
1.00 25.52
ATOM    615  NE  ARG  81     -3.063  22.554  20.439
1.00 29.78
ATOM    616  CZ  ARG  81     -2.811  22.897  19.179
1.00 30.17
ATOM    617  NH1 ARG  81     -3.810  22.847  18.270
1.00 32.23
ATOM    618  NH2 ARG  81     -1.547  23.168  18.822
1.00 30.78
ATOM    619  C   ARG  81     -7.071  20.736  24.659
1.00 13.57
ATOM    620  O   ARG  81     -8.067  20.129  24.261
1.00 12.68
ATOM    621  N   ASN  82     -7.100  21.628  25.667
1.00 13.97
ATOM    622  CA  ASN  82     -8.284  21.968  26.448
1.00 13.79
ATOM    623  CB  ASN  82     -9.250  22.793  25.588
1.00 14.31
ATOM    624  CG  ASN  82     -8.645  24.103  25.115
1.00 15.90
ATOM    625  OD1 ASN  82     -8.695  24.391  23.929
1.00 18.55
ATOM    626  ND2 ASN  82     -7.998  24.954  25.904
1.00 16.49
ATOM    627  C   ASN  82     -9.012  20.744  27.004
1.00 13.02
ATOM    628  O   ASN  82    -10.109  20.409  26.570
1.00 13.95
ATOM    629  N   PRO  83     -8.490  19.988  27.956
1.00 11.90
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ATOM    630  CD  PRO  83     -9.129  18.759  28.468
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ATOM    631  CA  PRO  83     -7.228  20.233  28.616
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ATOM    632  CB  PRO  83     -7.374  19.477  29.921
1.00 11.28
ATOM    633  CG  PRO  83     -8.168  18.254  29.514
1.00 12.43
ATOM    634  C   PRO  83     -6.020  19.812  27.806
1.00  9.55
ATOM    635  O   PRO  83     -6.065  18.925  26.941
1.00 10.93
ATOM    636  N   LEU  84     -4.949  20.487  28.139
1.00  9.55
ATOM    637  CA  LEU  84     -3.650  20.212  27.575
1.00 10.04
ATOM    638  CB  LEU  84     -2.725  21.373  27.908
1.00  9.09
ATOM    639  CG  LEU  84     -1.343  21.354  27.288
1.00  9.70
ATOM    640  CD1 LEU  84     -1.402  21.496  25.777
1.00  8.76
ATOM    641  CD2 LEU  84     -0.580  22.526  27.836
1.00 10.04
ATOM    642  C   LEU  84     -3.170  18.903  28.205
1.00  9.91
ATOM    643  O   LEU  84     -3.002  18.839  29.428
1.00 10.98
ATOM    644  N   ILE  85     -2.966  17.863  27.375
1.00 10.87
ATOM    645  CA  ILE  85     -2.538  16.538  27.873
1.00 11.93
ATOM    646  CB  ILE  85     -3.711  15.489  27.812
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ATOM    647  CG2 ILE  85     -3.226  14.128  28.360
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ATOM    648  CG1 ILE  85     -4.909  15.932  28.663
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ATOM    649  CD1 ILE  85     -4.578  16.047  30.178
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ATOM    650  C   ILE  85     -1.393  16.023  27.028
1.00 10.50
ATOM    651  O   ILE  85     -1.481  16.104  25.799
1.00 12.37
ATOM    652  N   GLU  86     -0.310  15.580  27.654
1.00 10.70
ATOM    653  CA  GLU  86      0.725  14.868  26.909
1.00 10.27
ATOM    654  CB  GLU  86      2.072  15.269  27.364
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ATOM    655  CG  GLU  86      3.207  14.580  26.675
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ATOM    656  CD  GLU  86      4.537  15.107  27.134
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ATOM    657  OE1 GLU  86      5.414  15.338  26.303
1.00  9.55
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ATOM    658  OE2 GLU    86       4.717  15.329  28.333
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ATOM    659  C   GLU    86       0.491  13.405  27.268
1.00  9.41
ATOM    660  O   GLU    86       0.386  13.100  28.464
1.00 10.26
ATOM    661  N   TYR    87       0.324  12.484  26.315
1.00  7.81
ATOM    662  CA  TYR    87       0.083  11.088  26.705
1.00  7.22
ATOM    663  CB  TYR    87      -1.380  10.679  26.368
1.00  5.93
ATOM    664  CG  TYR    87      -1.738  10.658  24.884
1.00  7.18
ATOM    665  CD1 TYR    87      -1.474   9.535  24.099
1.00  8.82
ATOM    666  CE1 TYR    87      -1.766   9.541  22.758
1.00  7.19
ATOM    667  CD2 TYR    87      -2.307  11.750  24.278
1.00  6.85
ATOM    668  CE2 TYR    87      -2.612  11.756  22.941
1.00  5.57
ATOM    669  CZ  TYR    87      -2.335  10.650  22.181
1.00  7.53
ATOM    670  OH  TYR    87      -2.601  10.636  20.820
1.00  7.92
ATOM    671  C   TYR    87       1.089  10.145  26.007
1.00  7.60
ATOM    672  O   TYR    87       1.593  10.460  24.914
1.00  6.22
ATOM    673  N   TYR    88       1.277   8.961  26.612
1.00  7.31
ATOM    674  CA  TYR    88       2.217   7.936  26.154
1.00  7.56
ATOM    675  CB  TYR    88       3.528   7.871  26.929
1.00  7.13
ATOM    676  CG  TYR    88       4.468   9.042  27.007
1.00  8.10
ATOM    677  CD1 TYR    88       4.116  10.234  27.638
1.00  7.71
ATOM    678  CE1 TYR    88       5.051  11.255  27.679
1.00  9.49
ATOM    679  CD2 TYR    88       5.718   8.892  26.443
1.00  9.02
ATOM    680  CE2 TYR    88       6.648   9.911  26.481
1.00  9.73
ATOM    681  CZ  TYR    88       6.293  11.080  27.099
1.00  8.95
ATOM    682  OH  TYR    88       7.201  12.087  27.106
1.00 10.51
ATOM    683  C   TYR    88       1.650   6.523  26.351
1.00  8.02
ATOM    684  O   TYR    88       1.044   6.170  27.392
1.00  9.02
ATOM    685  N   ILE    89       1.732   5.702  25.323
1.00  8.56
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ATOM    686  CA  ILE  89      1.416   4.327  25.590
1.00  8.75
ATOM    687  CB  ILE  89      0.027   3.839  24.820
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ATOM    688  CG2 ILE  89     -1.015   4.940  24.514
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ATOM    689  CG1 ILE  89      0.325   3.284  23.516
1.00 14.51
ATOM    690  CD1 ILE  89      0.188   1.799  23.814
1.00 14.55
ATOM    691  C   ILE  89      2.753   3.699  25.133
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ATOM    692  O   ILE  89      3.184   3.694  23.968
1.00  7.22
ATOM    693  N   VAL  90      3.532   3.345  26.134
1.00  5.27
ATOM    694  CA  VAL  90      4.871   2.849  25.887
1.00  7.56
ATOM    695  CB  VAL  90      5.825   3.471  27.042
1.00  8.16
ATOM    696  CG1 VAL  90      5.051   3.700  28.284
1.00 11.52
ATOM    697  CG2 VAL  90      7.017   2.585  27.388
1.00  9.08
ATOM    698  C   VAL  90      4.978   1.309  25.703
1.00  7.15
ATOM    699  O   VAL  90      4.665   0.486  26.563
1.00  5.67
ATOM    700  N   GLU  91      5.408   0.986  24.489
1.00  7.73
ATOM    701  CA  GLU  91      5.615  -0.376  24.005
1.00  6.96
ATOM    702  CB  GLU  91      5.524  -0.319  22.468
1.00  6.72
ATOM    703  CG  GLU  91      4.117   0.109  21.997
1.00  6.68
ATOM    704  CD  GLU  91      2.997  -0.826  22.456
1.00  7.56
ATOM    705  OE1 GLU  91      2.351  -0.578  23.452
1.00  6.34
ATOM    706  OE2 GLU  91      2.725  -1.823  21.810
1.00  7.24
ATOM    707  C   GLU  91      6.930  -1.017  24.472
1.00  8.03
ATOM    708  O   GLU  91      7.030  -2.224  24.753
1.00  8.58
ATOM    709  N   ASN  92      7.982  -0.202  24.624
1.00  6.40
ATOM    710  CA  ASN  92      9.301  -0.640  24.993
1.00  5.56
ATOM    711  CB  ASN  92      9.971  -1.242  23.777
1.00  7.20
ATOM    712  CG  ASN  92     11.086  -2.249  24.078
1.00  4.55
ATOM    713  OD1 ASN  92     11.508  -2.921  23.158
1.00  8.31
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ATOM    714  ND2 ASN    92      11.658   -2.485   25.235
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ATOM    715  C   ASN    92      10.101    0.567   25.492
1.00  6.87
ATOM    716  O   ASN    92       9.730    1.715   25.226
1.00  6.30
ATOM    717  N   PHE    93      11.178    0.358   26.247
1.00  6.43
ATOM    718  CA  PHE    93      12.014    1.435   26.793
1.00  7.21
ATOM    719  CB  PHE    93      11.316    2.114   27.994
1.00  5.14
ATOM    720  CG  PHE    93      10.879    1.272   29.174
1.00  5.75
ATOM    721  CD1 PHE    93      11.782    0.935   30.168
1.00  5.51
ATOM    722  CD2 PHE    93       9.551    0.889   29.272
1.00  5.97
ATOM    723  CE1 PHE    93      11.341    0.209   31.274
1.00  6.76
ATOM    724  CE2 PHE    93       9.115    0.163   30.383
1.00  6.85
ATOM    725  CZ  PHE    93      10.013   -0.176   31.378
1.00  6.45
ATOM    726  C   PHE    93      13.343    0.843   27.218
1.00  7.46
ATOM    727  O   PHE    93      13.439   -0.393   27.217
1.00  8.45
ATOM    728  N   GLY    94      14.326    1.659   27.611
1.00  9.27
ATOM    729  CA  GLY    94      15.674    1.230   27.935
1.00  9.67
ATOM    730  C   GLY    94      15.826    1.048   29.440
1.00 10.54
ATOM    731  O   GLY    94      14.938    0.537   30.111
1.00 11.63
ATOM    732  N   THR    95      16.920    1.556   29.966
1.00  8.02
ATOM    733  CA  THR    95      17.305    1.389   31.345
1.00  9.56
ATOM    734  CB  THR    95      18.812    1.747   31.485
1.00 10.30
ATOM    735  OG1 THR    95      18.950    3.144   31.160
1.00 12.79
ATOM    736  CG2 THR    95      19.687    0.913   30.581
1.00 11.07
ATOM    737  C   THR    95      16.493    2.215   32.339
1.00  9.93
ATOM    738  O   THR    95      16.646    2.037   33.545
1.00  7.89
ATOM    739  N   TYR    96      15.683    3.177   31.892
1.00  9.40
ATOM    740  CA  TYR    96      15.008    4.021   32.850
1.00  7.32
ATOM    741  CB  TYR    96      15.273    5.509   32.531
1.00  8.01
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ATOM    742  CG  TYR  96      14.506   6.456  33.440
1.00  8.66
ATOM    743  CD1 TYR  96      15.004   6.750  34.690
1.00 11.35
ATOM    744  CE1 TYR  96      14.312   7.595  35.549
1.00 12.60
ATOM    745  CD2 TYR  96      13.322   7.013  33.043
1.00  9.65
ATOM    746  CE2 TYR  96      12.629   7.862  33.892
1.00 12.25
ATOM    747  CZ  TYR  96      13.125   8.140  35.144
1.00 12.02
ATOM    748  OH  TYR  96      12.398   8.947  36.037
1.00 16.30
ATOM    749  C   TYR  96      13.547   3.703  32.772
1.00  7.36
ATOM    750  O   TYR  96      12.891   3.804  31.729
1.00  7.79
ATOM    751  N   ASN  97      13.023   3.350  33.925
1.00  6.69
ATOM    752  CA  ASN  97      11.637   2.945  33.955
1.00  8.46
ATOM    753  CB  ASN  97      11.378   2.061  35.197
1.00  6.22
ATOM    754  CG  ASN  97      10.028   1.337  35.187
1.00  8.71
ATOM    755  OD1 ASN  97       9.902   0.092  35.210
1.00  8.88
ATOM    756  ND2 ASN  97       8.950   2.081  35.177
1.00  4.75
ATOM    757  C   ASN  97      10.775   4.208  33.979
1.00  9.04
ATOM    758  O   ASN  97      10.891   5.009  34.918
1.00 10.16
ATOM    759  N   PRO  98       9.864   4.383  33.017
1.00  9.73
ATOM    760  CD  PRO  98       9.616   3.413  31.965
1.00 10.11
ATOM    761  CA  PRO  98       8.929   5.503  32.905
1.00 11.05
ATOM    762  CB  PRO  98       8.134   5.233  31.671
1.00 10.57
ATOM    763  CG  PRO  98       8.894   4.168  30.909
1.00 11.36
ATOM    764  C   PRO  98       7.999   5.746  34.094
1.00 12.84
ATOM    765  O   PRO  98       7.553   6.881  34.328
1.00 12.98
ATOM    766  N   SER  99       7.635   4.699  34.852
1.00 11.41
ATOM    767  CA  SER  99       6.808   4.887  36.009
1.00 12.40
ATOM    768  CB  SER  99       6.193   3.565  36.411
1.00 11.96
ATOM    769  OG  SER  99       7.223   2.731  36.896
1.00 13.35
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ATOM    770  C   SER  99      7.561   5.483  37.229
1.00 13.13
ATOM    771  O   SER  99      6.887   5.667  38.238
1.00 12.90
ATOM    772  N   THR 100      8.872   5.814  37.202
1.00 12.61
ATOM    773  CA  THR 100      9.570   6.319  38.373
1.00 14.31
ATOM    774  CB  THR 100     11.053   6.616  37.908
1.00 14.57
ATOM    775  OG1 THR 100     11.681   5.381  37.513
1.00 13.39
ATOM    776  CG2 THR 100     11.872   7.330  39.000
1.00 13.16
ATOM    777  C   THR 100      8.855   7.562  38.979
1.00 15.95
ATOM    778  O   THR 100      8.704   7.778  40.199
1.00 16.24
ATOM    779  N   GLY 101      8.283   8.459  38.197
1.00 16.98
ATOM    780  CA  GLY 101      7.638   9.603  38.908
1.00 22.81
ATOM    781  C   GLY 101      6.275   9.277  39.557
1.00 22.53
ATOM    782  O   GLY 101      5.846   9.799  40.603
1.00 24.30
ATOM    783  N   ALA 102      5.687   8.272  38.943
1.00 21.22
ATOM    784  CA  ALA 102      4.281   8.251  38.892
1.00 20.59
ATOM    785  CB  ALA 102      3.987   7.618  37.568
1.00 22.27
ATOM    786  C   ALA 102      3.361   7.726  39.918
1.00 20.55
ATOM    787  O   ALA 102      3.672   6.926  40.813
1.00 21.31
ATOM    788  N   THR 103      2.136   8.214  39.718
1.00 19.36
ATOM    789  CA  THR 103      1.096   7.726  40.572
1.00 18.69
ATOM    790  CB  THR 103      0.225   8.936  40.951
1.00 19.51
ATOM    791  OG1 THR 103      1.084   9.806  41.743
1.00 20.44
ATOM    792  CG2 THR 103     -1.075   8.498  41.648
1.00 18.11
ATOM    793  C   THR 103      0.371   6.668  39.775
1.00 16.70
ATOM    794  O   THR 103     -0.096   7.016  38.695
1.00 15.70
ATOM    795  N   LYS 104      0.313   5.418  40.253
1.00 15.07
ATOM    796  CA  LYS 104     -0.377   4.378  39.528
1.00 14.87
ATOM    797  CB  LYS 104      0.040   2.976  40.009
1.00 15.19
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ATOM    798  CG  LYS  104      -0.609   1.894  39.106
1.00 14.18
ATOM    799  CD  LYS  104      -0.270   0.443  39.507
1.00 13.52
ATOM    800  CE  LYS  104      -1.134  -0.507  38.694
1.00 13.51
ATOM    801  NZ  LYS  104      -0.693  -1.884  38.679
1.00 14.08
ATOM    802  C   LYS  104      -1.865   4.540  39.739
1.00 15.06
ATOM    803  O   LYS  104      -2.343   4.776  40.852
1.00 16.27
ATOM    804  N   LEU  105      -2.604   4.497  38.663
1.00 14.64
ATOM    805  CA  LEU  105      -4.012   4.642  38.765
1.00 16.10
ATOM    806  CB  LEU  105      -4.554   5.692  37.835
1.00 18.74
ATOM    807  CG  LEU  105      -4.239   7.181  38.163
1.00 21.39
ATOM    808  CD1 LEU  105      -2.801   7.479  37.844
1.00 21.27
ATOM    809  CD2 LEU  105      -5.016   8.118  37.214
1.00 22.30
ATOM    810  C   LEU  105      -4.735   3.403  38.415
1.00 17.33
ATOM    811  O   LEU  105      -5.958   3.403  38.613
1.00 18.71
ATOM    812  N   GLY  106      -4.142   2.420  37.716
1.00 16.78
ATOM    813  CA  GLY  106      -4.924   1.256  37.323
1.00 14.69
ATOM    814  C   GLY  106      -4.212   0.434  36.268
1.00 14.81
ATOM    815  O   GLY  106      -2.996   0.602  36.101
1.00 14.33
ATOM    816  N   GLU  107      -4.969  -0.468  35.612
1.00 14.31
ATOM    817  CA  GLU  107      -4.471  -1.347  34.563
1.00 14.01
ATOM    818  CB  GLU  107      -4.139  -2.724  35.141
1.00 17.27
ATOM    819  CG  GLU  107      -3.101  -2.650  36.288
1.00 19.43
ATOM    820  CD  GLU  107      -3.045  -3.847  37.242
1.00 21.27
ATOM    821  OE1 GLU  107      -3.493  -4.937  36.863
1.00 20.90
ATOM    822  OE2 GLU  107      -2.507  -3.677  38.362
1.00 22.15
ATOM    823  C   GLU  107      -5.503  -1.522  33.465
1.00 12.12
ATOM    824  O   GLU  107      -6.703  -1.267  33.664
1.00 11.04
ATOM    825  N   VAL  108      -5.003  -1.974  32.315
1.00 11.87
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ATOM    826  CA  VAL  108      -5.812  -2.265  31.129
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ATOM    827  CB  VAL  108      -6.069  -0.940  30.283
1.00 12.75
ATOM    828  CG1 VAL  108      -4.742  -0.406  29.730
1.00 13.07
ATOM    829  CG2 VAL  108      -6.925  -1.184  29.045
1.00 13.15
ATOM    830  C   VAL  108      -5.034  -3.299  30.306
1.00 11.86
ATOM    831  O   VAL  108      -3.796  -3.269  30.185
1.00 12.43
ATOM    832  N   THR  109      -5.786  -4.210  29.734
1.00 12.65
ATOM    833  CA  THR  109      -5.255  -5.257  28.889
1.00 12.30
ATOM    834  CB  THR  109      -5.935  -6.599  29.286
1.00 14.44
ATOM    835  OG1 THR  109      -5.674  -6.816  30.697
1.00 15.40
ATOM    836  CG2 THR  109      -5.415  -7.780  28.462
1.00 13.97
ATOM    837  C   THR  109      -5.579  -4.839  27.469
1.00 11.75
ATOM    838  O   THR  109      -6.711  -4.454  27.199
1.00 12.42
ATOM    839  N   SER  110      -4.630  -4.859  26.538
1.00 11.20
ATOM    840  CA  SER  110      -4.879  -4.532  25.155
1.00 10.80
ATOM    841  CB  SER  110      -4.941  -3.031  24.965
1.00 10.12
ATOM    842  OG  SER  110      -5.204  -2.682  23.632
1.00 11.48
ATOM    843  C   SER  110      -3.690  -5.105  24.405
1.00 11.32
ATOM    844  O   SER  110      -2.547  -5.207  24.920
1.00 12.50
ATOM    845  N   ASP  111      -4.042  -5.590  23.213
1.00 10.79
ATOM    846  CA  ASP  111      -3.119  -6.087  22.199
1.00 12.25
ATOM    847  CB  ASP  111      -2.447  -4.890  21.486
1.00 13.12
ATOM    848  CG  ASP  111      -3.520  -3.986  20.911
1.00 13.55
ATOM    849  OD1 ASP  111      -4.270  -4.400  20.041
1.00 13.05
ATOM    850  OD2 ASP  111      -3.631  -2.862  21.377
1.00 14.27
ATOM    851  C   ASP  111      -2.044  -7.029  22.706
1.00 13.33
ATOM    852  O   ASP  111      -0.825  -6.910  22.477
1.00 14.68
ATOM    853  N   GLY  112      -2.491  -7.933  23.566
1.00 13.67
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ATOM    854  CA  GLY  112     -1.595  -8.956  24.072
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ATOM    855  C   GLY  112     -0.959  -8.664  25.418
1.00  9.91
ATOM    856  O   GLY  112     -0.094  -9.437  25.783
1.00  9.84
ATOM    857  N   SER  113     -1.218  -7.600  26.180
1.00  9.51
ATOM    858  CA  SER  113     -0.518  -7.452  27.424
1.00  8.66
ATOM    859  CB  SER  113      0.851  -6.812  27.189
1.00  6.40
ATOM    860  OG  SER  113      1.712  -6.818  28.313
1.00  5.69
ATOM    861  C   SER  113     -1.350  -6.588  28.292
1.00  8.88
ATOM    862  O   SER  113     -2.213  -5.874  27.803
1.00 11.46
ATOM    863  N   VAL  114     -1.190  -6.691  29.594
1.00 10.48
ATOM    864  CA  VAL  114     -1.787  -5.641  30.395
1.00  9.89
ATOM    865  CB  VAL  114     -2.304  -6.209  31.826
1.00 11.18
ATOM    866  CG1 VAL  114     -1.870  -7.641  32.032
1.00 10.85
ATOM    867  CG2 VAL  114     -1.853  -5.324  32.981
1.00 10.22
ATOM    868  C   VAL  114     -0.666  -4.581  30.466
1.00 10.04
ATOM    869  O   VAL  114      0.535  -4.838  30.231
1.00  9.80
ATOM    870  N   TYR  115     -1.145  -3.368  30.683
1.00  9.29
ATOM    871  CA  TYR  115     -0.390  -2.144  30.797
1.00  9.53
ATOM    872  CB  TYR  115     -0.789  -1.113  29.736
1.00  8.60
ATOM    873  CG  TYR  115     -0.417  -1.372  28.281
1.00  8.75
ATOM    874  CD1 TYR  115      0.505  -0.554  27.607
1.00  7.26
ATOM    875  CE1 TYR  115      0.788  -0.783  26.275
1.00  7.31
ATOM    876  CD2 TYR  115     -1.043  -2.408  27.631
1.00  8.71
ATOM    877  CE2 TYR  115     -0.778  -2.646  26.312
1.00  9.10
ATOM    878  CZ  TYR  115      0.126  -1.843  25.640
1.00  8.99
ATOM    879  OH  TYR  115      0.311  -2.161  24.309
1.00  9.97
ATOM    880  C   TYR  115     -0.747  -1.545  32.151
1.00 10.03
ATOM    881  O   TYR  115     -1.874  -1.667  32.641
1.00 10.65
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ATOM    882  N   ASP 116      0.206  -0.895  32.794
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ATOM    883  CA  ASP 116     -0.094  -0.187  34.027
1.00 10.95
ATOM    884  CB  ASP 116      1.092  -0.386  34.982
1.00 10.88
ATOM    885  CG  ASP 116      1.153  -1.800  35.539
1.00 12.59
ATOM    886  OD1 ASP 116      2.085  -2.539  35.245
1.00 12.09
ATOM    887  OD2 ASP 116      0.281  -2.189  36.298
1.00 13.31
ATOM    888  C   ASP 116     -0.323   1.296  33.610
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ATOM    889  O   ASP 116      0.307   1.792  32.656
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ATOM    890  N   ILE 117     -1.259   1.980  34.272
1.00 10.31
ATOM    891  CA  ILE 117     -1.768   3.320  33.944
1.00 11.24
ATOM    892  CB  ILE 117     -3.316   3.226  33.891
1.00 11.80
ATOM    893  CG2 ILE 117     -3.969   4.626  33.919
1.00 12.99
ATOM    894  CG1 ILE 117     -3.714   2.409  32.671
1.00 11.14
ATOM    895  CD1 ILE 117     -5.195   2.011  32.763
1.00 11.30
ATOM    896  C   ILE 117     -1.287   4.285  35.037
1.00 11.21
ATOM    897  O   ILE 117     -1.484   4.007  36.236
1.00 11.16
ATOM    898  N   TYR 118     -0.646   5.368  34.631
1.00  9.67
ATOM    899  CA  TYR 118     -0.039   6.351  35.530
1.00 10.44
ATOM    900  CB  TYR 118      1.481   6.226  35.585
1.00  8.83
ATOM    901  CG  TYR 118      1.994   4.842  35.959
1.00  9.45
ATOM    902  CD1 TYR 118      2.210   3.919  34.944
1.00  9.25
ATOM    903  CE1 TYR 118      2.696   2.663  35.241
1.00 10.55
ATOM    904  CD2 TYR 118      2.261   4.524  37.284
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ATOM    905  CE2 TYR 118      2.756   3.269  37.601
1.00 11.19
ATOM    906  CZ  TYR 118      2.969   2.341  36.566
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ATOM    907  OH  TYR 118      3.484   1.078  36.857
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ATOM    908  C   TYR 118     -0.329   7.801  35.103
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ATOM    909  O   TYR 118     -0.521   8.073  33.914
1.00  9.48
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ATOM    910  N   ARG  119      -0.335   8.724  36.053
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ATOM    911  CA  ARG  119      -0.553  10.155  35.779
1.00 14.61
ATOM    912  CB  ARG  119      -1.973  10.621  36.265
1.00 17.40
ATOM    913  CG  ARG  119      -2.089  12.021  36.952
1.00 23.37
ATOM    914  CD  ARG  119      -3.471  12.483  37.490
1.00 26.18
ATOM    915  NE  ARG  119      -4.356  12.883  36.371
1.00 30.66
ATOM    916  CZ  ARG  119      -5.263  13.922  36.488
1.00 33.03
ATOM    917  NH1 ARG  119      -5.325  14.574  37.682
1.00 34.14
ATOM    918  NH2 ARG  119      -6.085  14.337  35.453
1.00 32.49
ATOM    919  C   ARG  119       0.546  10.899  36.536
1.00 14.29
ATOM    920  O   ARG  119       0.866  10.575  37.684
1.00 12.19
ATOM    921  N   THR  120       1.155  11.881  35.873
1.00 15.13
ATOM    922  CA  THR  120       2.243  12.707  36.395
1.00 16.87
ATOM    923  CB  THR  120       3.620  12.350  35.730
1.00 19.15
ATOM    924  OG1 THR  120       3.353  12.150  34.313
1.00 22.72
ATOM    925  CG2 THR  120       4.277  11.062  36.276
1.00 21.25
ATOM    926  C   THR  120       1.859  14.152  35.996
1.00 15.72
ATOM    927  O   THR  120       0.915  14.327  35.236
1.00 11.42
ATOM    928  N   GLN  121       2.510  15.213  36.480
1.00 18.27
ATOM    929  CA  GLN  121       2.180  16.591  36.057
1.00 18.87
ATOM    930  CB  GLN  121       1.490  17.333  37.150
1.00 20.46
ATOM    931  CG  GLN  121       0.818  18.647  36.686
1.00 22.49
ATOM    932  CD  GLN  121       0.528  19.470  37.910
1.00 23.15
ATOM    933  OE1 GLN  121      -0.609  19.866  38.201
1.00 24.23
ATOM    934  NE2 GLN  121       1.566  19.744  38.711
1.00 25.37
ATOM    935  C   GLN  121       3.469  17.311  35.724
1.00 17.45
ATOM    936  O   GLN  121       4.483  17.265  36.429
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ATOM    937  N   ARG  122       3.507  17.884  34.557
1.00 16.86
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ATOM    938  CA  ARG  122      4.666  18.646  34.205
1.00 17.38
ATOM    939  CB  ARG  122      4.919  18.431  32.738
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ATOM    940  CG  ARG  122      4.983  16.936  32.371
1.00 17.66
ATOM    941  CD  ARG  122      6.035  16.268  33.234
1.00 19.91
ATOM    942  NE  ARG  122      7.405  16.749  33.155
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ATOM    943  CZ  ARG  122      8.060  17.199  32.061
1.00 21.20
ATOM    944  NH1 ARG  122      7.485  17.275  30.834
1.00 24.32
ATOM    945  NH2 ARG  122      9.359  17.539  32.223
1.00 21.52
ATOM    946  C   ARG  122      4.285  20.099  34.550
1.00 18.00
ATOM    947  O   ARG  122      3.127  20.529  34.309
1.00 15.92
ATOM    948  N   VAL  123      5.264  20.778  35.187
1.00 17.31
ATOM    949  CA  VAL  123      5.100  22.174  35.581
1.00 17.49
ATOM    950  CB  VAL  123      5.204  22.292  37.118
1.00 18.68
ATOM    951  CG1 VAL  123      5.057  23.752  37.578
1.00 18.13
ATOM    952  CG2 VAL  123      4.025  21.495  37.756
1.00 17.08
ATOM    953  C   VAL  123      6.132  23.037  34.888
1.00 18.01
ATOM    954  O   VAL  123      7.348  22.820  34.956
1.00 19.01
ATOM    955  N   ASN  124      5.550  23.905  34.058
1.00 18.77
ATOM    956  CA  ASN  124      6.246  24.856  33.204
1.00 18.46
ATOM    957  CB  ASN  124      6.839  25.978  34.049
1.00 18.90
ATOM    958  CG  ASN  124      5.688  26.871  34.516
1.00 19.83
ATOM    959  OD1 ASN  124      5.841  27.521  35.548
1.00 21.50
ATOM    960  ND2 ASN  124      4.457  26.916  33.966
1.00 18.83
ATOM    961  C   ASN  124      7.288  24.274  32.301
1.00 17.97
ATOM    962  O   ASN  124      8.485  24.580  32.249
1.00 19.21
ATOM    963  N   GLN  125      6.718  23.436  31.457
1.00 17.42
ATOM    964  CA  GLN  125      7.513  22.692  30.522
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ATOM    965  CB  GLN  125      7.340  21.162  30.781
1.00 18.71
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ATOM    967  CD  GLN  125      9.279  20.949  32.484
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ATOM    968  OE1 GLN  125     10.114  20.190  31.977
1.00 24.89
ATOM    969  NE2 GLN  125      9.737  21.872  33.335
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ATOM    970  C   GLN  125      6.994  23.068  29.153
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ATOM    971  O   GLN  125      5.828  23.466  28.999
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ATOM    972  N   PRO  126      7.847  22.921  28.138
1.00 13.24
ATOM    973  CD  PRO  126      9.304  22.804  28.272
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ATOM    974  CA  PRO  126      7.444  22.936  26.773
1.00 11.98
ATOM    975  CB  PRO  126      8.686  22.587  25.972
1.00 12.53
ATOM    976  CG  PRO  126      9.718  22.098  26.983
1.00 12.93
ATOM    977  C   PRO  126      6.291  22.004  26.506
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ATOM    978  O   PRO  126      6.115  20.913  27.073
1.00 11.75
ATOM    979  N   SER  127      5.462  22.567  25.667
1.00 11.03
ATOM    980  CA  SER  127      4.315  21.866  25.199
1.00 10.71
ATOM    981  CB  SER  127      3.081  22.230  26.043
1.00 10.39
ATOM    982  OG  SER  127      2.618  23.555  25.769
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ATOM    983  C   SER  127      4.178  22.399  23.787
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ATOM    984  O   SER  127      4.895  23.295  23.342
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ATOM    985  N   ILE  128      3.221  21.771  23.145
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ATOM    986  CA  ILE  128      2.628  22.112  21.877
1.00 12.75
ATOM    987  CB  ILE  128      1.518  20.968  21.777
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ATOM    988  CG2 ILE  128      0.710  20.959  23.097
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ATOM    989  CG1 ILE  128      0.526  21.141  20.704
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ATOM    990  CD1 ILE  128     -0.807  20.429  21.073
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ATOM    991  C   ILE  128      2.166  23.616  21.786
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ATOM    992  O   ILE  128      2.010  24.160  20.689
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ATOM    993  N   ILE  129      1.911  24.268  22.923
1.00 11.55
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ATOM    995  CB  ILE  129      0.050  25.264  23.942  1.00 12.77
ATOM    996  CG2 ILE  129      0.063  25.810  25.372  1.00 12.19
ATOM    997  CG1 ILE  129     -1.129  25.730  23.208  1.00 13.88
ATOM    998  CD1 ILE  129     -1.556  24.732  22.166  1.00 13.59
ATOM    999  C   ILE  129      2.414  26.584  23.628  1.00 12.93
ATOM   1000  O   ILE  129      2.195  27.795  23.818  1.00 15.68
ATOM   1001  N   GLY  130      3.595  26.061  24.001  1.00 12.43
ATOM   1002  CA  GLY  130      4.647  26.814  24.669  1.00 11.72
ATOM   1003  C   GLY  130      4.752  26.447  26.146  1.00 10.71
ATOM   1004  O   GLY  130      4.105  25.502  26.558  1.00 11.85
ATOM   1005  N   THR  131      5.552  27.094  26.999  1.00 10.63
ATOM   1006  CA  THR  131      5.633  26.774  28.422  1.00 10.32
ATOM   1007  CB  THR  131      6.507  27.778  29.134  1.00 10.53
ATOM   1008  OG1 THR  131      7.632  27.883  28.294  1.00 13.71
ATOM   1009  CG2 THR  131      6.969  27.382  30.523  1.00  9.71
ATOM   1010  C   THR  131      4.284  26.743  29.108  1.00 10.05
ATOM   1011  O   THR  131      3.512  27.720  29.121  1.00  9.35
ATOM   1012  N   ALA  132      4.066  25.560  29.667  1.00  9.69
ATOM   1013  CA  ALA  132      2.783  25.208  30.228  1.00 10.33
ATOM   1014  CB  ALA  132      1.913  24.574  29.134  1.00  8.89
ATOM   1015  C   ALA  132      2.850  24.229  31.413  1.00 10.62
ATOM   1016  O   ALA  132      3.926  23.711  31.762  1.00 11.81
ATOM   1017  N   THR  133      1.721  24.123  32.100  1.00 11.01
ATOM   1018  CA  THR  133      1.541  23.113  33.134  1.00 12.77
ATOM   1019  CB  THR  133      1.145  23.789  34.478  1.00 13.67
ATOM   1020  OG1 THR  133      2.360  24.447  34.897  1.00 15.23
ATOM   1021  CG2 THR  133      0.651  22.835  35.592  1.00 13.85
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ATOM   1022  C   THR 133      0.457  22.171  32.632  1.00 12.16
ATOM   1023  O   THR 133     -0.628  22.577  32.176  1.00 12.10
ATOM   1024  N   PHE 134      0.737  20.871  32.678  1.00 12.04
ATOM   1025  CA  PHE 134     -0.191  19.880  32.118  1.00 10.26
ATOM   1026  CB  PHE 134      0.014  19.753  30.593  1.00  9.74
ATOM   1027  CG  PHE 134      1.450  19.497  30.158  1.00 10.10
ATOM   1028  CD1 PHE 134      2.345  20.538  30.021  1.00  9.16
ATOM   1029  CD2 PHE 134      1.867  18.209  29.908  1.00  9.83
ATOM   1030  CE1 PHE 134      3.652  20.313  29.647  1.00 10.74
ATOM   1031  CE2 PHE 134      3.177  17.977  29.532  1.00 10.39
ATOM   1032  CZ  PHE 134      4.074  19.013  29.401  1.00 11.34
ATOM   1033  C   PHE 134      0.017  18.520  32.763  1.00 10.97
ATOM   1034  O   PHE 134      1.072  18.261  33.334  1.00  9.19
ATOM   1035  N   TYR 135     -1.010  17.700  32.729  1.00 10.76
ATOM   1036  CA  TYR 135     -0.896  16.327  33.212  1.00 11.10
ATOM   1037  CB  TYR 135     -2.247  15.786  33.699  1.00 11.88
ATOM   1038  CG  TYR 135     -2.575  16.298  35.090  1.00 15.19
ATOM   1039  CD1 TYR 135     -1.816  15.890  36.171  1.00 17.24
ATOM   1040  CE1 TYR 135     -2.054  16.351  37.457  1.00 17.77
ATOM   1041  CD2 TYR 135     -3.607  17.183  35.298  1.00 17.57
ATOM   1042  CE2 TYR 135     -3.862  17.655  36.594  1.00 18.71
ATOM   1043  CZ  TYR 135     -3.082  17.237  37.661  1.00 18.40
ATOM   1044  OH  TYR 135     -3.300  17.720  38.949  1.00 21.11
ATOM   1045  C   TYR 135     -0.420  15.448  32.070  1.00 11.69
ATOM   1046  O   TYR 135     -0.785  15.621  30.892  1.00  9.54
ATOM   1047  N   GLN 136      0.343  14.419  32.467  1.00 10.85
ATOM   1048  CA  GLN 136      0.927  13.498  31.514  1.00 10.50
ATOM   1049  CB  GLN 136      2.404  13.687  31.779  1.00 10.66
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ATOM   1050  CG  GLN  136      3.383  13.299  30.739
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ATOM   1051  CD  GLN  136      4.796  13.457  31.286
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ATOM   1052  OE1 GLN  136      5.081  13.041  32.405
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ATOM   1053  NE2 GLN  136      5.779  13.990  30.586
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ATOM   1054  C   GLN  136      0.324  12.109  31.805
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ATOM   1055  O   GLN  136      0.386  11.631  32.935
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ATOM   1056  N   TYR  137     -0.377  11.467  30.878
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ATOM   1057  CA  TYR  137     -0.971  10.159  31.163
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ATOM   1058  CB  TYR  137     -2.397  10.001  30.623
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ATOM   1059  CG  TYR  137     -3.416  11.066  30.980
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ATOM   1060  CD1 TYR  137     -3.310  11.813  32.136
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ATOM   1061  CE1 TYR  137     -4.280  12.773  32.443
1.00 22.42
ATOM   1062  CD2 TYR  137     -4.483  11.262  30.111
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ATOM   1063  CE2 TYR  137     -5.460  12.203  30.402
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ATOM   1064  CZ  TYR  137     -5.354  12.951  31.583
1.00 23.68
ATOM   1065  OH  TYR  137     -6.404  13.809  31.984
1.00 26.30
ATOM   1066  C   TYR  137     -0.113   9.085  30.487
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ATOM   1067  O   TYR  137      0.402   9.271  29.361
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ATOM   1068  N   TRP  138      0.043   7.923  31.137
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ATOM   1069  CA  TRP  138      0.942   6.879  30.640
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ATOM   1070  CB  TRP  138      2.273   6.810  31.444
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ATOM   1071  CG  TRP  138      3.238   8.005  31.558
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ATOM   1072  CD2 TRP  138      4.474   8.150  30.977
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ATOM   1073  CE2 TRP  138      4.909   9.361  31.532
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ATOM   1074  CE3 TRP  138      5.287   7.474  30.094
1.00  7.01
ATOM   1075  CD1 TRP  138      2.949   9.072  32.391
1.00  9.63
ATOM   1076  NE1 TRP  138      3.986   9.872  32.348
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ATOM   1077  CZ2 TRP  138      6.134   9.919  31.237
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ATOM   1078  CZ3 TRP   138      6.516   8.026  29.791
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ATOM   1079  CH2 TRP   138      6.935   9.224  30.349
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ATOM   1080  C   TRP   138      0.270   5.502  30.768
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ATOM   1081  O   TRP   138     -0.484   5.232  31.709
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ATOM   1082  N   SER   139      0.417   4.664  29.751
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ATOM   1083  CA  SER   139      0.116   3.213  29.806
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ATOM   1084  CB  SER   139     -0.762   2.735  28.692
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ATOM   1087  O   SER   139      2.063   2.824  28.486
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ATOM   1088  N   VAL   140      2.027   1.780  30.460
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ATOM   1089  CA  VAL   140      3.306   1.139  30.256
1.00  9.29
ATOM   1090  CB  VAL   140      4.191   1.381  31.514
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ATOM   1091  CG1 VAL   140      5.580   0.791  31.315
1.00  9.67
ATOM   1092  CG2 VAL   140      4.329   2.918  31.750
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ATOM   1093  C   VAL   140      3.057  -0.352  30.005
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ATOM   1094  O   VAL   140      2.461  -1.081  30.830
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ATOM   1095  N   ARG   141      3.521  -0.782  28.835
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ATOM   1096  CA  ARG   141      3.404  -2.199  28.443
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ATOM   1097  CB  ARG   141      3.757  -2.391  26.945
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ATOM   1098  CG  ARG   141      3.431  -3.843  26.522
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ATOM   1099  CD  ARG   141      3.684  -4.280  25.085
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ATOM   1100  NE  ARG   141      2.351  -4.384  24.581
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ATOM   1101  CZ  ARG   141      1.620  -5.350  24.013
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ATOM   1102  NH1 ARG   141      1.970  -6.616  23.698
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ATOM   1103  NH2 ARG   141      0.361  -4.935  23.852
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ATOM   1104  C   ARG   141      4.314  -3.116  29.302
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ATOM   1105  O   ARG   141      5.544  -2.998  29.343
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ATOM   1106  N   ARG  142      3.671  -4.076  30.002  1.00 10.19
ATOM   1107  CA  ARG  142      4.421  -5.006  30.855  1.00 10.06
ATOM   1108  CB  ARG  142      3.456  -5.843  31.649  1.00  9.21
ATOM   1109  CG  ARG  142      2.846  -5.061  32.777  1.00  9.22
ATOM   1110  CD  ARG  142      2.254  -5.997  33.780  1.00  8.97
ATOM   1111  NE  ARG  142      1.440  -5.256  34.689  1.00  8.70
ATOM   1112  CZ  ARG  142      0.517  -5.842  35.463  1.00  9.47
ATOM   1113  NH1 ARG  142      0.338  -7.202  35.422  1.00  9.32
ATOM   1114  NH2 ARG  142     -0.249  -5.019  36.194  1.00  9.02
ATOM   1115  C   ARG  142      5.337  -5.888  30.030  1.00  9.64
ATOM   1116  O   ARG  142      6.540  -5.988  30.280  1.00 11.41
ATOM   1117  N   ASN  143      4.786  -6.349  28.929  1.00  9.44
ATOM   1118  CA  ASN  143      5.490  -7.214  27.997  1.00 11.07
ATOM   1119  CB  ASN  143      4.523  -8.194  27.420  1.00 15.18
ATOM   1120  CG  ASN  143      4.798  -9.487  28.077  1.00 19.83
ATOM   1121  OD1 ASN  143      4.354  -9.693  29.224  1.00 21.43
ATOM   1122  ND2 ASN  143      5.589 -10.295  27.336  1.00 21.58
ATOM   1123  C   ASN  143      6.111  -6.441  26.874  1.00  9.46
ATOM   1124  O   ASN  143      5.418  -6.281  25.886  1.00  7.69
ATOM   1125  N   HIS  144      7.379  -6.024  26.936  1.00  8.08
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ATOM   1127  CB  HIS  144      9.257  -4.577  26.418  1.00  9.04
ATOM   1128  CG  HIS  144      9.304  -3.876  27.793  1.00 10.00
ATOM   1129  CD2 HIS  144     10.469  -3.373  28.345  1.00  9.60
ATOM   1130  ND1 HIS  144      8.353  -3.647  28.706  1.00 12.26
ATOM   1131  CE1 HIS  144      8.904  -3.055  29.762  1.00 10.72
ATOM   1132  NE2 HIS  144     10.191  -2.893  29.533  1.00  9.65
ATOM   1133  C   HIS  144      8.017  -5.649  24.551  1.00 11.38
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ATOM   1136  CA  ARG  145      7.940  -5.277  22.118
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ATOM   1137  CB  ARG  145      6.587  -5.753  21.574
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ATOM   1138  CG  ARG  145      5.436  -4.936  22.132
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ATOM   1139  CD  ARG  145      4.240  -4.414  21.384
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ATOM   1143  NH2 ARG  145      1.255  -5.917  20.002
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ATOM   1144  C   ARG  145      8.377  -4.136  21.243
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ATOM   1145  O   ARG  145      8.135  -2.979  21.592
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ATOM   1153  CA  SER  147      6.519  -4.300  16.562
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ATOM   1154  CB  SER  147      7.027  -5.161  15.395
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ATOM   1157  O   SER  147      5.045  -5.529  17.975
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ATOM   1159  CA  GLY  148      2.718  -4.855  16.790
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ATOM   1164  CB  SER 149     -1.780  -3.739  16.037  1.00 12.62
ATOM   1165  OG  SER 149     -1.203  -4.402  14.925  1.00 20.18
ATOM   1166  C   SER 149     -1.039  -2.529  18.002  1.00 11.07
ATOM   1167  O   SER 149     -0.827  -3.223  19.016  1.00 10.99
ATOM   1168  N   VAL 150     -1.551  -1.292  18.042  1.00 10.64
ATOM   1169  CA  VAL 150     -2.081  -0.728  19.273  1.00 10.28
ATOM   1170  CB  VAL 150     -1.224   0.457  19.813  1.00 10.28
ATOM   1171  CG1 VAL 150     -1.893   1.108  20.981  1.00  8.88
ATOM   1172  CG2 VAL 150      0.152  -0.067  20.216  1.00 10.04
ATOM   1173  C   VAL 150     -3.475  -0.217  18.919  1.00 11.39
ATOM   1174  O   VAL 150     -3.657   0.510  17.936  1.00 11.90
ATOM   1175  N   ASN 151     -4.456  -0.661  19.661  1.00 11.61
ATOM   1176  CA  ASN 151     -5.782  -0.144  19.539  1.00 13.38
ATOM   1177  CB  ASN 151     -6.745  -1.312  19.809  1.00 15.35
ATOM   1178  CG  ASN 151     -8.191  -0.831  19.729  1.00 19.10
ATOM   1179  OD1 ASN 151     -8.500   0.361  19.505  1.00 21.76
ATOM   1180  ND2 ASN 151     -9.156  -1.690  20.022  1.00 19.23
ATOM   1181  C   ASN 151     -5.885   1.011  20.561  1.00 13.32
ATOM   1182  O   ASN 151     -6.102   0.814  21.778  1.00 13.06
ATOM   1183  N   THR 152     -5.744   2.285  20.109  1.00 12.70
ATOM   1184  CA  THR 152     -5.670   3.429  21.041  1.00 10.80
ATOM   1185  CB  THR 152     -5.402   4.741  20.273  1.00  9.08
ATOM   1186  OG1 THR 152     -6.506   5.026  19.423  1.00  9.66
ATOM   1187  CG2 THR 152     -4.107   4.646  19.511  1.00  8.63
ATOM   1188  C   THR 152     -6.908   3.610  21.922  1.00 12.10
ATOM   1189  O   THR 152     -6.824   3.983  23.109  1.00 12.41
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ATOM	1191	CA	ALA	153	-9.365	3.291	22.021
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ATOM	1192	CB	ALA	153	-10.422	2.571	21.212
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ATOM	1193	C	ALA	153	-9.390	2.663	23.410
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ATOM	1194	O	ALA	153	-9.868	3.237	24.389
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ATOM	1195	N	ASN	154	-8.688	1.526	23.512
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ATOM	1196	CA	ASN	154	-8.642	0.814	24.781
1.00	13.58						
ATOM	1197	CB	ASN	154	-7.924	-0.499	24.613
1.00	14.46						
ATOM	1198	CG	ASN	154	-8.774	-1.450	23.787
1.00	15.29						
ATOM	1199	OD1	ASN	154	-8.242	-2.299	23.070
1.00	16.38						
ATOM	1200	ND2	ASN	154	-10.098	-1.363	23.743
1.00	15.71						
ATOM	1201	C	ASN	154	-7.982	1.583	25.887
1.00	13.14						
ATOM	1202	O	ASN	154	-8.449	1.613	27.026
1.00	12.59						
ATOM	1203	N	HIS	155	-6.889	2.234	25.498
1.00	12.89						
ATOM	1204	CA	HIS	155	-6.134	3.043	26.440
1.00	13.28						
ATOM	1205	CB	HIS	155	-4.802	3.434	25.858
1.00	12.80						
ATOM	1206	CG	HIS	155	-3.934	2.210	25.718
1.00	11.42						
ATOM	1207	CD2	HIS	155	-3.836	1.416	24.591
1.00	10.81						
ATOM	1208	ND1	HIS	155	-3.200	1.700	26.674
1.00	9.77						
ATOM	1209	CE1	HIS	155	-2.651	0.622	26.199
1.00	9.97						
ATOM	1210	NE2	HIS	155	-3.033	0.454	24.962
1.00	11.54						
ATOM	1211	C	HIS	155	-6.908	4.318	26.755
1.00	13.91						
ATOM	1212	O	HIS	155	-7.062	4.658	27.932
1.00	14.83						
ATOM	1213	N	PHE	156	-7.439	5.021	25.752
1.00	14.01						
ATOM	1214	CA	PHE	156	-8.141	6.257	26.072
1.00	14.84						
ATOM	1215	CB	PHE	156	-8.592	6.997	24.827
1.00	14.21						
ATOM	1216	CG	PHE	156	-7.473	7.369	23.896
1.00	14.75						
ATOM	1217	CD1	PHE	156	-6.201	7.640	24.371
1.00	16.50						

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ATOM   1219  CE1 PHE   156      -5.187   7.970  23.480
1.00 16.11
ATOM   1220  CE2 PHE   156      -6.705   7.762  21.658
1.00 15.84
ATOM   1221  CZ  PHE   156      -5.435   8.039  22.124
1.00 15.88
ATOM   1222  C   PHE   156      -9.382   5.954  26.906
1.00 15.07
ATOM   1223  O   PHE   156      -9.769   6.777  27.733
1.00 14.39
ATOM   1224  N   ASN   157      -9.998   4.775  26.741
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ATOM   1225  CA  ASN   157     -11.188   4.463  27.487
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ATOM   1226  CB  ASN   157     -11.947   3.247  26.911
1.00 19.91
ATOM   1227  CG  ASN   157     -12.692   3.666  25.658
1.00 21.97
ATOM   1228  OD1 ASN   157     -12.844   2.871  24.723
1.00 23.02
ATOM   1229  ND2 ASN   157     -13.152   4.918  25.484
1.00 22.77
ATOM   1230  C   ASN   157     -10.865   4.175  28.900
1.00 16.15
ATOM   1231  O   ASN   157     -11.579   4.674  29.773
1.00 16.04
ATOM   1232  N   ALA   158      -9.781   3.426  29.138
1.00 16.31
ATOM   1233  CA  ALA   158      -9.319   3.129  30.494
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ATOM   1234  CB  ALA   158      -8.057   2.270  30.451
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ATOM   1235  C   ALA   158      -8.968   4.411  31.265
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ATOM   1236  O   ALA   158      -9.334   4.645  32.446
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ATOM   1237  N   TRP   159      -8.223   5.257  30.544
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ATOM   1238  CA  TRP   159      -7.821   6.552  31.114
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ATOM   1239  CB  TRP   159      -6.966   7.314  30.077
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ATOM   1240  CG  TRP   159      -5.581   6.715  29.752
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ATOM   1241  CD2 TRP   159      -4.763   7.058  28.689
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ATOM   1242  CE2 TRP   159      -3.698   6.146  28.818
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ATOM   1243  CE3 TRP   159      -4.769   7.967  27.653
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ATOM   1244  CD1 TRP   159      -5.023   5.673  30.479
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ATOM   1245  NE1 TRP   159      -3.886   5.350  29.897
1.00 14.73
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ATOM 1246 CZ2 TRP 159 -2.648 6.129 27.922 1.00 12.93
ATOM 1247 CZ3 TRP 159 -3.718 7.949 26.765 1.00 14.15
ATOM 1248 CH2 TRP 159 -2.678 7.043 26.901 1.00 14.30
ATOM 1249 C TRP 159 -9.063 7.367 31.526 1.00 14.08
ATOM 1250 O TRP 159 -9.173 7.852 32.677 1.00 13.42
ATOM 1251 N ALA 160 -10.062 7.392 30.651 1.00 13.53
ATOM 1252 CA ALA 160 -11.307 8.092 30.906 1.00 15.69
ATOM 1253 CB ALA 160 -12.248 7.868 29.726 1.00 16.17
ATOM 1254 C ALA 160 -12.001 7.663 32.188 1.00 17.30
ATOM 1255 O ALA 160 -12.385 8.505 33.015 1.00 16.76
ATOM 1256 N SER 161 -12.076 6.354 32.474 1.00 19.40
ATOM 1257 CA SER 161 -12.799 5.887 33.642 1.00 21.24
ATOM 1258 CB SER 161 -13.287 4.489 33.431 1.00 21.88
ATOM 1259 OG SER 161 -12.407 3.847 32.515 1.00 24.44
ATOM 1260 C SER 161 -11.964 5.918 34.877 1.00 21.71
ATOM 1261 O SER 161 -12.392 5.514 35.946 1.00 24.49
ATOM 1262 N HIS 162 -10.712 6.282 34.759 1.00 23.09
ATOM 1263 CA HIS 162 -9.899 6.476 35.932 1.00 24.28
ATOM 1264 CB HIS 162 -8.462 5.957 35.756 1.00 26.20
ATOM 1265 CG HIS 162 -8.372 4.525 36.263 1.00 27.93
ATOM 1266 CD2 HIS 162 -8.372 3.431 35.418 1.00 29.19
ATOM 1267 ND1 HIS 162 -8.356 4.063 37.523 1.00 27.81
ATOM 1268 CE1 HIS 162 -8.362 2.734 37.460 1.00 29.60
ATOM 1269 NE2 HIS 162 -8.366 2.358 36.196 1.00 30.08
ATOM 1270 C HIS 162 -9.870 7.965 36.145 1.00 24.43
ATOM 1271 O HIS 162 -9.187 8.427 37.059 1.00 25.01
ATOM 1272 N GLY 163 -10.594 8.702 35.302 1.00 24.39
ATOM 1273 CA GLY 163 -10.836 10.108 35.509 1.00 24.50

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ATOM   1275  O   GLY  163     -9.693  12.118  35.003
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ATOM   1276  N   LEU  164     -9.231  10.416  33.695
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ATOM   1277  CA  LEU  164     -8.277  11.191  32.910
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ATOM   1278  CB  LEU  164     -7.044  10.365  32.732
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ATOM   1279  CG  LEU  164     -5.870  10.394  33.748
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ATOM   1280  CD1 LEU  164     -6.313  10.578  35.218
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ATOM   1281  CD2 LEU  164     -5.109   9.096  33.505
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ATOM   1282  C   LEU  164     -8.824  11.631  31.569
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ATOM   1284  N   THR  165     -9.577  12.737  31.598
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ATOM   1285  CA  THR  165    -10.232  13.318  30.432
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ATOM   1286  CB  THR  165    -11.074  14.530  30.985
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ATOM   1287  OG1 THR  165    -11.760  15.097  29.861
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ATOM   1292  CA  LEU  166     -8.746  14.319  26.983
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ATOM   1296  CD2 LEU  166     -6.070  12.560  26.494
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ATOM   1298  O   LEU  166    -10.797  15.041  26.120
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ATOM   1301  C   GLY  167     -9.629  17.078  23.482
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ATOM   1304  CA  THR 168     -9.390  17.997  21.191
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ATOM   1305  CB  THR 168     -9.760  19.453  20.735
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ATOM   1311  CA  MET 169     -6.576  15.814  19.662
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ATOM   1312  CB  MET 169     -6.836  14.483  18.935
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ATOM   1313  CG  MET 169     -7.727  13.517  19.684
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ATOM   1314  SD  MET 169     -7.005  13.060  21.275
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ATOM   1315  CE  MET 169     -6.344  11.447  20.893
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ATOM   1316  C   MET 169     -5.637  16.648  18.797
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ATOM   1317  O   MET 169     -6.024  17.252  17.775
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ATOM   1318  N   ASP 170     -4.409  16.773  19.220
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ATOM   1319  CA  ASP 170     -3.401  17.330  18.307
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ATOM   1320  C   ASP 170     -2.670  16.197  17.581
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ATOM   1321  O   ASP 170     -3.383  15.283  17.156
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ATOM   1324  OD1 ASP 170     -2.595  20.457  18.060
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ATOM   1326  N   TYR 171     -1.371  16.150  17.418
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ATOM   1327  CA  TYR 171     -0.722  15.072  16.692
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ATOM   1328  CB  TYR 171      0.658  15.560  16.280
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ATOM   1329  CG  TYR 171      1.776  15.763  17.280
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ATOM	1331	CE1	TYR	171	3.502	14.868	18.710	1.00 11.22
ATOM	1332	CD2	TYR	171	2.214	17.053	17.525	1.00 10.25
ATOM	1333	CE2	TYR	171	3.298	17.256	18.365	1.00 11.78
ATOM	1334	CZ	TYR	171	3.942	16.171	18.954	1.00 10.98
ATOM	1335	OH	TYR	171	5.018	16.389	19.792	1.00 9.29
ATOM	1336	C	TYR	171	-0.664	13.748	17.485	1.00 11.01
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ATOM	1338	N	GLN	172	-0.495	12.610	16.787	1.00 10.87
ATOM	1339	CA	GLN	172	-0.503	11.246	17.332	1.00 9.53
ATOM	1340	CB	GLN	172	-1.889	10.738	17.176	1.00 10.18
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ATOM	1343	OE1	GLN	172	-4.267	8.939	16.379	1.00 10.99
ATOM	1344	NE2	GLN	172	-4.460	8.919	18.599	1.00 9.74
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ATOM	1346	O	GLN	172	0.248	10.353	15.235	1.00 9.11
ATOM	1347	N	ILE	173	1.605	10.159	17.064	1.00 6.96
ATOM	1348	CA	ILE	173	2.851	9.711	16.474	1.00 6.99
ATOM	1349	CB	ILE	173	3.719	11.020	16.654	1.00 9.21
ATOM	1350	CG2	ILE	173	4.913	11.022	17.588	1.00 8.02
ATOM	1351	CG1	ILE	173	4.058	11.318	15.266	1.00 10.65
ATOM	1352	CD1	ILE	173	2.845	12.113	14.728	1.00 11.75
ATOM	1353	C	ILE	173	3.439	8.426	17.059	1.00 5.19
ATOM	1354	O	ILE	173	3.197	8.136	18.232	1.00 5.97
ATOM	1355	N	VAL	174	4.168	7.610	16.319	1.00 5.30
ATOM	1356	CA	VAL	174	4.915	6.536	16.939	1.00 5.81
ATOM	1357	CB	VAL	174	5.054	5.263	16.052	1.00 5.74

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ATOM   1359  CG2 VAL   174      3.714   4.576  15.922
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ATOM   1360  C   VAL   174      6.265   7.190  17.103
1.00  4.60
ATOM   1361  O   VAL   174      6.897   7.637  16.138
1.00  6.58
ATOM   1362  N   ALA   175      6.711   7.294  18.342
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ATOM   1363  CA  ALA   175      7.916   8.061  18.607
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ATOM   1367  N   VAL   176     10.275   7.682  18.999
1.00  3.82
ATOM   1368  CA  VAL   176     11.309   7.278  19.902
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ATOM   1369  CB  VAL   176     12.544   6.510  19.224
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ATOM   1370  CG1 VAL   176     11.996   5.545  18.180
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ATOM   1371  CG2 VAL   176     13.562   7.396  18.645
1.00 10.56
ATOM   1372  C   VAL   176     11.770   8.574  20.585
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ATOM   1373  O   VAL   176     11.913   9.639  19.970
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ATOM   1375  CA  GLU   177     12.231   9.635  22.760
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ATOM   1377  CG  GLU   177     11.585  10.946  24.816
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ATOM   1379  OE1 GLU   177     10.780  12.349  26.527
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ATOM   1380  OE2 GLU   177      9.286  11.451  25.259
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ATOM   1381  C   GLU   177     13.517   9.156  23.393
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ATOM   1388  CA  TYR  179      17.120  11.392  27.288
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ATOM   1389  CB  TYR  179      16.265  11.279  28.515
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ATOM   1390  CG  TYR  179      16.568  12.436  29.468
1.00 12.04
ATOM   1391  CD1 TYR  179      17.399  12.215  30.546
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ATOM   1392  CE1 TYR  179      17.723  13.240  31.395
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ATOM   1393  CD2 TYR  179      16.046  13.693  29.239
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ATOM   1394  CE2 TYR  179      16.363  14.734  30.096
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ATOM   1395  CZ  TYR  179      17.202  14.485  31.159
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ATOM   1400  CA  PHE  180      20.844  11.857  27.265
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ATOM   1401  CB  PHE  180      21.070  12.542  28.622
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ATOM   1402  CG  PHE  180      20.877  14.060  28.563
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ATOM   1403  CD1 PHE  180      19.621  14.613  28.705
1.00  9.46
ATOM   1404  CD2 PHE  180      21.983  14.868  28.362
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ATOM   1405  CE1 PHE  180      19.455  15.988  28.651
1.00 10.39
ATOM   1406  CE2 PHE  180      21.829  16.257  28.304
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ATOM   1407  CZ  PHE  180      20.563  16.793  28.449
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ATOM   1408  C   PHE  180      21.464  10.460  27.223
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ATOM   1409  O   PHE  180      22.029   9.956  28.189
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ATOM   1410  N   SER  181      21.345   9.832  26.067
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ATOM   1411  CA  SER  181      21.791   8.442  25.903
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ATOM   1412  CB  SER  181      20.605   7.545  26.290
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ATOM   1413  OG  SER  181      19.481   7.785  25.454
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ATOM   1415  O   SER  181      22.630   9.127  23.718
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ATOM   1416  N   SER  182      22.284   6.908  24.101
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ATOM   1417  CA  SER  182      22.600   6.513  22.769
1.00  9.70
ATOM   1418  CB  SER  182      24.021   6.016  22.725
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ATOM   1419  OG  SER  182      24.934   7.051  23.106
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ATOM   1420  C   SER  182      21.630   5.414  22.351
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ATOM   1421  O   SER  182      20.985   4.798  23.214
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ATOM   1422  N   GLY  183      21.467   5.216  21.045
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ATOM   1423  CA  GLY  183      20.679   4.114  20.539
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ATOM   1424  C   GLY  183      20.553   4.164  19.022
1.00 11.09
ATOM   1425  O   GLY  183      21.209   4.956  18.319
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ATOM   1426  N   SER  184      19.610   3.365  18.544
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ATOM   1427  CA  SER  184      19.353   3.151  17.133
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ATOM   1428  CB  SER  184      20.242   2.035  16.632
1.00 11.41
ATOM   1429  OG  SER  184      20.580   2.124  15.275
1.00 15.69
ATOM   1430  C   SER  184      17.911   2.693  17.021
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ATOM   1432  N   ALA  185      17.201   3.136  16.005
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ATOM   1434  CB  ALA  185      14.812   3.279  16.605
1.00  5.28
ATOM   1435  C   ALA  185      15.548   2.893  14.307
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ATOM   1436  O   ALA  185      15.852   3.971  13.757
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ATOM   1437  N   SER  186      14.890   1.908  13.712
1.00  8.21
ATOM   1438  CA  SER  186      14.334   2.079  12.414
1.00  7.45
ATOM   1439  CB  SER  186      15.201   1.275  11.479
1.00  8.66
ATOM   1440  OG  SER  186      14.672   1.319  10.175
1.00  8.84
ATOM   1441  C   SER  186      12.886   1.622  12.514
1.00  9.02
```

```
ATOM   1442  O   SER  186      12.620   0.477  12.885
1.00  7.60
ATOM   1443  N   ILE  187      11.875   2.493  12.354
1.00  8.36
ATOM   1444  CA  ILE  187      10.517   2.013  12.471
1.00  9.26
ATOM   1445  CB  ILE  187       9.933   2.345  13.963
1.00 12.57
ATOM   1446  CG2 ILE  187      11.001   2.794  14.977
1.00 10.66
ATOM   1447  CG1 ILE  187       8.893   3.361  13.908
1.00 11.46
ATOM   1448  CD1 ILE  187       7.616   2.580  14.124
1.00 12.68
ATOM   1449  C   ILE  187       9.623   2.516  11.337
1.00  9.78
ATOM   1450  O   ILE  187       9.873   3.551  10.672
1.00 10.18
ATOM   1451  N   THR  188       8.595   1.714  11.054
1.00  7.46
ATOM   1452  CA  THR  188       7.617   2.008  10.005
1.00  8.18
ATOM   1453  CB  THR  188       7.753   0.972   8.796
1.00  6.41
ATOM   1454  OG1 THR  188       9.078   1.119   8.291
1.00  5.82
ATOM   1455  CG2 THR  188       6.704   1.165   7.675
1.00  5.98
ATOM   1456  C   THR  188       6.218   1.951  10.610
1.00  6.88
ATOM   1457  O   THR  188       5.864   1.006  11.312
1.00  7.67
ATOM   1458  N   VAL  189       5.464   2.989  10.331
1.00  7.06
ATOM   1459  CA  VAL  189       4.104   3.196  10.803
1.00  9.10
ATOM   1460  CB  VAL  189       3.913   4.657  11.335
1.00  9.32
ATOM   1461  CG1 VAL  189       2.554   4.764  11.989
1.00  9.82
ATOM   1462  CG2 VAL  189       4.969   5.015  12.359
1.00  7.64
ATOM   1463  C   VAL  189       3.135   2.976   9.664
1.00  8.74
ATOM   1464  O   VAL  189       3.432   3.326   8.514
1.00  9.63
ATOM   1465  N   SER  190       1.967   2.498  10.032
1.00 10.99
ATOM   1466  CA  SER  190       0.853   2.346   9.138
1.00 13.10
ATOM   1467  CB  SER  190       0.966   1.065   8.258
1.00 15.33
ATOM   1468  OG  SER  190       1.169  -0.182   8.908
1.00 15.84
ATOM   1469  C   SER  190      -0.415   2.271   9.950
1.00 14.43
```

ATOM 1470 O SER 190 -1.439 2.580 9.354 1.00 17.68
ATOM 1471 OXT SER 190 -0.392 1.975 11.150 1.00 12.62

A portion of the three-dimensional crystal structure of a mutant of the *B. circulans* xylanase containing an intramolecular disulfide bond is shown in Figure 4. In this mutant serine 100 and asparagine 148 have been mutated to cysteine. The disulfide which is formed links the last strand of sheet III to the alpha-helix. The structure f this mutant is identical to the wild-type enzyme except for the side chains of residues 100 and 148 and some very minor shifts of nearby atoms. The same region of the superimposed structures of the *B. circulans* and *T. harzianum* xylanases is shown in Figure 5. Further details of the analyses will be providedin the examples, however, one can see that although there are some sequence differences between the *B. circulans* and *T. harzianum* xylanases near the site of the manufactured disulfide and some small position differences at other nearby residues, the structure of the *T. harzianum* xylanase is virtually identical to the structure of the *B. circulans* xylanase at the positions of the mutations. The sequence alignment (Figure 1) shows that the sequence homology of the various xylanases in the vicinity of residues 98 to 100 and 148 to 152 is very good. One would therefore expect that the structures of other related xylanases would be similar enough that they could be modified with the introduction of an intramolecular disulfide bond linking sheet III to the alpha-helix and that this disulfide bond would stabilize them in a simliar manner as taught in the present invention.

Similarly one would expect that sequence changes made at the N-terminal end of the *B. circulans* xylanase, which produced a more stable xylanase, could be mad at corresponding positions in other family G xylanases, with a reasonable expectation of success. For example, the

strategy to substitute the Asn residue N-8 of *B. circulans* xylanase by Tyr and Phe to yield more stable mutants, can be readily applied to fungal xylanases of *A. niger var. awamori*, *A. tubigensis A* and *T. ressei I*. These fungal xylanses, like the bacterial *B. circulans* xylanase, possesses the same target Asn residue, available for the same mutation.

For some xylanases with reduced sequence homology to the *B. circulans* xylanase it may be necessary to make other mutations to increase the homology in the vicinity of the introduced intramolecular disulfide bond or N-terminal mutation. In other words it may be necessary to make another xylanase more like the *B. circulans* xylanase before the disulfide bond could be formed successfully or before a useful N-terminal mutation is made. Throughout the application reference is made to amino acid positions based on the *B. circulans* xylanase, as a reference. Figure 1 is provided in order to determine the corresponding position in other family G xylanases.

In the modification of family G xylanases, the *B. circulans* xylanase (BCX) was chosen to exemplify the principles of the present invention. This enzyme is only moderately thermostable at temperatures up to 55°C. For application of this enzyme in pretreatment of kraft-pulp to enhance bleaching, a higher temperature stability is desirable. As noted previously, a xylanase which has a higher pH optimum, would also be benificial in the bleaching step. Such a modified enzyme would also be useful in the food industry.

As noted previously, the chief application of the modified xylanase is for pulp biobleaching in the production of paper. Before bleaching process, the pulp is hot at a temperature range of 55-70°C and in an alkalin

state. Many commercial wild-type xylanases (*T. r esee* and *B. pummilus*) only function at 55°C and some require acidic pH. Therefore the desirable improvements in a mutant xylanase are higher functional temperature (temperature optimum) and pH (pH optimum), with the former characteristic as the most important. Xylanase of higher temperature optimum can be used to treat pulp at high temperature without idle period of cooling. With higher functional pH (pH optimal), the mutant enzyme would requir less or no acid to neutralize the pulp which is basic at that stage. The economic benefits include saving in time, acid for neutralization and no acidic corrosion of the equipments. Therefore a desirable xylanase mutant would have a temperature optimum approaching 70°C and a pH optimum approaching 9 in pulp bleaching. Of course any measurable improvement over the wild type xylanase, BCX (54°C, pH 7) would be beneficial.

In the parent application (U.S. Serial No. 08/044,621), the improvement of mutant was measured in terms of thermostability, ie, its ability to survive incubation at high temperature in buffer without any substrate. Increased thermostability means an increase of 2°C to 15°C, while maintaining at least 60% of activity, compared to wild type xylanase, after incubation at an elevated temperature. Such a pursuit of improv d thermostability or passive heat-resistance was the sole objective in another publication involving xylanase (Arase et al, 1993). Although this characteristic may be useful and possibly related to the shelf-life of the enzyme, it is n t a reliable indicator of the ability of the enzyme to function at that temperature. At the elevated temperature, the enzyme molecule may have been deactivated or unfolded but managed to be quickly refolded or reactivated before assayed at 40°C. Therefor the gain in th rmostability or heat-resistance ($T_{1/2}$) does not guarantee that the enzyme remains intact or can function at higher temperature.

An alternative assay was used to determine if the mutant enzyme can function at high temperature. Instead of incubation in buffer without xylan, xylanase is incubated in the same manner with xylan in the buffer. The modified process is designed to evaluate the hydrolysis of soluble xylan by the mutant enzyme at different temperatures. The reducing sugar released was then determined by the HBAH reagent. With addition of xylan in the mixture, the protocol remained essentially the same. The temperature optimum is defined as the highest temperature at which 150% relative enzymatic activity was maintained as compared to performance at 40°C.

A further measure of increased stability, according to the present invention, is an increase in pH optimum in the hydrolysis of xylan. In the present invention, pH optimum has been defined as the highest pH at which the enzyme still maintains 50% of the maximal activity.

In addition to the assay based on the direct hydrolysis of xylan, the mutant xylanases were also tested for their ability to bleach pulp. The temperature optimum and pH optimum in pulp bleaching are defined as the highest temperature and pH at which the brightness gain by the treated pulp was not less than 0.4.

In one aspect of the present invention, the starting point was the solution of the three-dimensional structure of the enzyme (Figure 2). By inspection of the structure, a prediction of the residues, which could be mutated to cysteine, and which would possibly oxidize to form an intra-molecular SS bridge was made. This was accomplished by searching for pairs of residues for which the inter-C-alpha distance was less than 6.6 Å and the inter-C-beta distance was less than 4.5 Å. If either member of the pair was a glycine this pair was ignored

because glycines are important for the backbone conformation. Some candidates were excluded because they were involved in the active site of the enzyme. In addition the distance algorithm had selected pairs of residues which were too close, as they were on adjacent strands of one of the beta sheets.

Two areas for the introduction of a cysteine residue were selected. These areas are the beta-sheet III and the alpha-helix. Although the alpha-helix itself ranges from amino acids 147 to 155, some residues on either side could potentially be used to form intra-molecular disulfide linkages. Accordingly, the amino acids in the alpha-helix ranging from 143 to 158 and the amino acids on the beta-sheet III ranging from 95 to 109 are potential sites for the introduction of cysteine residues for the formation of an intra-molecular disulphide bond between these two areas. Two pairs of residues were found (details will be provided in the Examples) and the corresponding mutants were constructed, and in both cases the intra-molecular SS bond formed spontaneously.

In addition to the intra-molecular SS bond mutant, a second type of disulfide mutant was constructed by joining two protein molecules with an inter-molecular SS bond. For the production of this type of disulfide bond, a cysteine is introduced in each of the two xylanase protein molecules such that the cysteine is on the exterior of the molecule, thus the two molecules can be joined by an inter-molecular disulphide bond. Potential sites of interest include amino acid 15 to 31 of the beta-sheet I; amino acid 43 to 61 of the beta-sheet I; amino acid 87 to 104 of the loop and beta-sheet III; amino acid 133 to 163 of the alpha-helix and surrounding loops; and amino acids 177 to 185 of the beta-sheet I.

A third type of mutant unrelated to SS bond formation has also been found to confer stability. These mutants were generated by specific mutagenesis of amino acids at the N-terminus of the xylanase molecule. In one embodiment of the present invention, amino acids 1 to 25 of the xylanase protein are selected for specific mutagenesis. As discussed above, the amino acid numbering is based on the *B. circulans* xylanase. Corresponding amino acids from other family G xylanases can be determined by making reference to Figure 1.

In one example of the present invention, the introduction of tyrosine or phenylalanine by substitution resulted in mutants with increased thermostability, temperature optimum and pH optimum, when compared to the wild type protein. In another embodiment of the present invention, the introduction of proline at amino acid position 22, by substitution resulted in a more stable mutant. In a further example of the present invention, the stability of the mutant could be increased by combining two or mutations in a single molecule. In some examples the effect of two or more mutants was additive and in other examples the effect was co-operative.

The selected amino acids referred to above ar based on the amino acid sequence of the *B. circulans* xylanase sequence. The present invention is not limited to producing a thermostable xylanase from this bacterial source, but includes the production of a thermostable xylanase for other sources, as listed in Table 1 for example. The choice of suitable amino acids to target for mutagenesis in xylanases from other sources will be obvious to those skilled in the art by comparing the sequences and choosing the amino acids corresponding to those identified for the *B. circulans* xylanase.

The present invention will be further illustrated by way of the following examples, which are not to be construed as limiting. In these examples the xylanase mutants, which were constructed, are summarized in Table 2.

Table 2

Xylanase Mutants

Name	Mutation	Type of Mutation
TS1	S100C N148C	intra-molecular SS bond
TS2	V98C A152C	intra-molecular SS bond
TS3a	D4Y N8F	N-terminal Mutation
TS3	S100C N148C D4Y N8F	intra-molecular SS bond + N-terminal
TS4a	S179C dimer	inter-molecular SS bond
TS4	S100C N148C S179C monomer/dimer mix	intra- + inter-molecular SS bond
TS4M	S100C N148C S179C monomer	intra-molecular SS bond
TS4D	S100C N148C S179C dimer	intra- + inter-molecular SS bond
TS5a	N8Y	N-terminal mutation
TS6a	T3G D4Y N8Y	N-terminal mutation

EXAMPLE 1

CONSTRUCTION OF THE MUTANT TS1

The gene encoding the *B. subtilis* xylanase (see Figure 6), is identical to the *B. circulans* xylanase except at position 147, where a serine (BSX) rather than threonine (BCX) is encoded, (Yang et al, 1988, Nucleic Acid Research 16:7187 and Paice et al, 1986, Archives of Microbiology 144:201-206; all references incorporated herein by reference). The xylanase gene encoded by pBSX was mutated by the uracil containing DNA (UDNA) method (Kunkle et al, 1987, Methods in Enzymology, 154:367-382, which is incorporated herein by reference) to produce the mutant S100C. The coding sequence for this mutant gene was then removed from the vector using PCR. The 5′ portion of the resulting PCR product (codons 1-103) was combined with a synthetic gene fragment from the *B. circulans* gene (codons 104-185) to produce the plasmid pCWBCX::S100C. The region containing codons 136-153 of pCWBCX::S100C was replaced with synthetic oligonucleotides to introduce the second cysteine codon at position 148 (N148C). The resultant plasmid contained the S100C and N148C mutations and was called TS1.

All liquid cultures in this and other Examples were grown in either 2YT medium (16 g yeast extract, 10 g bacto-tryptone, 5 g NaCl, 1 L of H_2O), or TB medium (24 g yeast extract, 12 g bacto-tryptone, 10 ml 1 M potassium phosphate buffer pH 7.5, 5 ml of 80% glycerol, 1 L H_2O). The antibiotic ampicillin was added at 150 µg/ml to all cultures of plasmid containing strains. The cultures were grown with shaking at 30°C for protein and plasmid production, and 37°C for the production of single stranded DNA containing particles.

Basic recombinant DNA methods like plasmid DNA isolation, restriction enzyme digestions, the purification of DNA fragments for cloning, ligations, transformations and DNA sequencing were performed as recommended by the enzyme supplier, or the manufacturer of the kit us d for

the particular procedure. Polyacrylamide gel electrophoresis of proteins was performed as recommended in the technical literature supplied by Bio-Rad laboratories, Mississauga Ont. Restriction and DNA modification enzymes were purchased from New England Biolabs LTD., Mississauga Ont. Prep-A-Gene DNA purification matrix was purchased from Bio-Rad laboratories, Mississauga Ont. Sequenase, a DNA sequencing kit, was purchased from US Biochemicals, Cleveland Ohio. Oligonucleotide 3' end labelling was performed with a kit from Boehringer Mannheim Canada, Laval PQ. Protein concentration was determined from the molar extinction coefficient of the xylanase: 81,790 $L.mol^{-1}$.

A 2 ml culture of *Escherichia coli* RZ1032 (HfrKL16PO/45 [lysA(61-62)], dut1, ung1, thi1, relA1, Zbd-279::Tn10,supE44) harbouring pBSX was grown at 37°C with vigourous shaking until the A_{600} reached 0.5, at which time 10 µl of the helper phage, M13KO7, (titre 1 x 10^{12}/ml), was added. After 1 h, 0.5 ml of the culture was subcultured into 20 ml of fresh media containing 50 µg kanamycin/ml, and 100 µg ampicillin/ml. This culture was shaken at 200 rpm for 16 h at 37°C. The supernatant containing the single stranded DNA containing particles (SSDNAP) was collected after centrifugation of the culture at 4°C for 20 min. at 7000 x g. The SSDNAP were precipitated by the addition of 1/4 volume of 15% PEG-8000/ 14.6% NaCl. After 30 min. at room temperature the precipitate was collected by centrifugation at 7000 x g for 20 min. The precipitate was resuspended in 0.5 ml TE buffer (10mM Tris - HCl, 1 mM EDTA, pH 8), and left on ice for 30 min. Any insoluble material was removed by a brief centrifugation in a microcentrifuge. The supernatant was made up to 1% acetic acid and the resultant precipitate was collected on a 1 cm glass fibre filter by suction filtration. Protein was removed from the precipitated SSDNAP'S, and the DNA bound to the filter by washing the filter under vacuum, with 2 ml of 4 M $NaClO_4$ in TE buffer. Excess $NaClO_4$ was removed by

washing the filter with 2 ml of ice cold 70% ethanol. The filter was allowed to air dry for 5 min. at room temperature. The filter was placed into a 500 µl microcentrifuge tube with a hole in the bottom. This tube was placed inside a 1.5 ml microcentrifuge tube. The single stranded DNA was eluted by the addition of 50µl of 0.1X TE buffer to the filter and after 5 min centrifuging the two tubes to recover the liquid in the larger tube. The elution step was repeated. The single stranded DNA was quantified by analysis on agarose gel electrophoresis.

Oligonucleotides were synthesized using an Applied Biosystems model 380B DNA synthesizer. Synthetic oligonucleotides were purified by polyacrylamide gel electrophoresis (PAGE) in 15% gels containing 7 M urea. Oligonucleotides were detected by UV shadowing, cut out and eluted from the gel slices in 1 ml of 0.5 M ammonium acetate, 10 mM magnesium acetate. The oligonucleotides were desalted by passage through a Sep-Pak C18 reverse phase cartridge (Atkinson and Smith, 1984, In: Oligonucleotide synthesis a practical approach. Gait, N. J., editor. IRL press, Washington). The oligonucleotides were eluted with 20 % acetonitrile, and the concentration was determined by reading the A_{260} of the solution. For the production of S100C the following oligonucleotide was synthesized:

SEQ ID NO:4

Codons 96 97 98 99 100 101 102 103 104

5′- GGT ACT GTA AAA TGT GAT GGG GGT ACA - 3′

The oligonucleotide was phosphorylated for use in mutagenesis. 100 to 200 pmol of the oligonucleotide was mixed with 2µl of 10X kinase buffer (500 mM Tris-HCl, pH 7.6, 100 mM $MgCl_2$, 100 mM dithiothreitol (DTT), 1.0 mM spermidine, 10 mM adenosine triphosphate (ATP)), and th volume was adjusted with water to a final volume of 20 µl.

The reaction was initiated by the addition of 10U of T4 polynucleotide kinase. The reaction was performed at 37°C for 1 h, after which the reaction was terminated by heating at 70°C for 10 min. Oligonucleotides prepared in this fashion were ready for use in the UDNA mutagenesis method.

The phosphorylated mutagenic oligonucleotide primer (20 pmol, 2μl), was mixed with the ssUDNA (0.5 pmol, 7 μl); 1 μl of 10X DNA synthesis buffer (200 mM Tris-HCl, pH 7.5, 100 mM $MgCl_2$, 500 mM NaCl, 10 mM DTT). The DNA's were annealed by heating the mixture to 70°C, and then slowly cooled to room temperature. The mutagenesis reaction was then performed by the addition of 10 μl of the DNA synthesis mixture (1 μl 10X synthesis buffer (200 mM Tris-HCl, pH 7.5, 100 mM $MgCl_2$, 100 mM DTT), 1 μl 5 mM deoxynucleotide triphosphates (dNTPs), 1 μl 10 mM ATP, 6 μl H_2O, 1 μl, 5U, T4 DNA ligase, 0.25 μl, 2.5U, T7 DNA polymerase). The reaction mixture was kept on ice for 5 min, then at room temperature for 5 min., and finally incubated at 37°C for 60 to 90 min. The reaction was terminated by the addition of 3 μl 50 mM EDTA. The reaction products were analyzed by standard agarose gel electrophoresis (Sambrook et al, 1989, Molecular cloning: A laboratory manual, 2nd edition). A 5 μl portion of this mixture was used to transform the *E. coli* strain MV1190 (deletion (lac-proAB), thi, supE44, deletion (srl-recA)306::Tn10(tet^r) [F':traD36, proAB, lacI^{q}deletion M15]) or BHM 71-18 (deletion (lac-proAB), thi, supE44, [mutS::Tn10(tet^r)]).

After transformation the bacteria were plated onto solid media (2YT) containing 150 μg ampicillin/ml. Individual colonies were picked and transferred to a fresh plate in a grid pattern of 100 colonies per plate. After overnight growth at 37°C, the colonies were transferred to Hybond-N nylon filter membranes (Amersham, Canada, Oakville Ont.), the colonies were lysed, and the DNA fixed to these

filters by well established procedures (Sambrook et al, 1989, Molecular cloning: a laboratory manual).

Oligonucleotide probes were synthesized using a 3′ end labelling kit from Boehringer Mannheim. 100 pmol of oligonucleotide were dissolved in 4μl H_2O. The reaction mixture was completed as follows: 4 μl of 5X tailing buffer (1M potassium cacodylate, 0.125 M Tris-HCl, bovine serum albumin (BSA) 1.25 mg/ml, pH 6.6), 4 μl 25 mM $CoCl_2$, 1 μl 1 mM digoxigenin-dideoxy-uracil triphosphate (DIG-ddUTP), 6 μl H_2O, 1 μl terminal transferase (50U). The mixture was reacted at 37°C for 15 min., then 2 μl of glycogen was added (0.2 μg / μl in 0.2 M EDTA). The labelled oligonucleotide was precipitated by the addition of 2.5 μl of 4 M LiCl, and 75 μl of ice cold, -20°C, 95% ethanol. After 2 h at - 20°C the precipitate was collected by centrifugation at 12,000 g for 30 min. The pellet was washed with 100 μl ice cold 70% ethanol, and then air dried in a fume hood.

The filters containing the colonies were washed free of cell debris in 2X SSC (SSC buffer is 0.15 M NaCl, 0.015 M sodium citrate, pH 7.0), 0.1% SDS, by gently rubbing the surface of the filter with a wet tissue. After removal of the cell debris, the filters were pre-hybridized in hybridization buffer (5X SSC, 0.1% SDS, 0.02% N-laurylsarkosine, 1% blocking reagent, a commercial product from Boehringer Mannheim), for at least 5 h. After the pre-hybridization, the DIG-labelled oligonucleotide was dissolved in 100 μl H_2O, and added to 7 ml of hybridization buffer. The hybridization buffer-DIG labelled oligonucleotide mixture was poured onto the filter and the hybridization was then performed in plastic petri dishes at 37°C, with shaking overnight. Excess (unbound) probe was removed by washing the filters twice for 10 min, each time, with 2X SSC,0.1% SDS at room temperature. Non-specifically

bound probe was removed by washing the filters at 40°C with 0.5 X SSC, 0.1% SDS, again twice for 10 min each time.

The filters were then washed in 25 ml of M buffer (0.1 M maleic acid, 0.15 M NaCl, pH adjusted to 7.5 with concentrated NaOH). The filter was then incubated in 20 ml M buffer containing 1% blocking reagent for 30 min. at room temperature. The solution was then discarded and then further incubated with 10 ml of fresh M buffer with 1% blocking reagent and 750 mU of anti-DIG-alkaline phosphatase-conjugate antibody. After 30 min., the filter was washed with 3 x 20 ml washes of M buffer with 0.3% Tween 20. The filter was then equilibrated in alkalin phosphatase buffer (0.1 M Tris-HCl, 0.1 M NaCl, 0.05 M $MgCl_2$ pH 9.5). The filter was then dipped in a solution of Lumi-phos, a chemiluminescent alkaline phosphatase substrate (Boehringer Mannheim). The filter was sealed into a plastic bag, and incubated at 37°C for 30 min. The filter in the bag was then exposed to x-ray film for 5 to 60 min. Positive colonies were further characterized by DNA sequencing. The clone chosen for subsequent work contained plasmid pBSXS100C.

Removal of the mature coding sequence of the *B. subtilis* xylanase gene from its leader sequence was accomplished using PCR with specific primers. The primers were designed so that the resulting amplification product would contain a new NdeI restriction site to allow the precise fusion of the coding region to the transcriptional signals present in pCWori+. An XbaI site was introduced at the 3′ end to facilitate cloning into pCWori+.

The plasmid pBSXS100C was digested with EcoRI to linearize it, and then it was used as a template for PCR. The amplification reaction contained, 15 µl of template DNA (50 ng), 5 µl of 10X buffer (100 mM KCl, 100 mM ammonium sulfate, 200 mM Tris-HCl pH 8.8, 40 mM magnesium sulfate,

1% Triton X100, 100 μg/ml BSA), 5 μl of 5 mM dNTPs, 2.5 μl 5′ primer solution (25 pmol), 2.5 μl 3′ primer solution (25 pmol), 19 μl H_2O, 1 μl (1U) Vent DNA polymerase (New England Biolabs). The 5′ and 3′ primers are shown below:

SEQ ID NO:5

NdeI site

5' BSYSL GC CTG CAG CAT ATG GCT AGC ACA GAC TAC TGG CAA AAT TGG A

SEQ ID NO:6

XbaI site

3' BSXYL GC AAG CTT TCT AGA CTT TAA CCA TTA CTA ACG ATT TTA ATA ATC

The reaction was covered with 50 μl of paraffin oil to prevent evaporation. The reaction mixture was prewarmed to 94°C without enzyme for 5 min., then the reaction mixture was cooled to 72°C, then the enzyme was added. The reaction was incubated in a temperature cycler for 30 cycles of 94°C 1 min., 55°C 2 min., 72°C for 2 min. The yield of amplification product was approximately 1μg of a 700 bp fragment. This fragment was purified from an agarose gel.

The resulting PCR product had an introduced NdeI site at the 5′ end to provide a start codon for the xylanase gene when cloned into pCWori+. The xylanase gene was cloned into pCWori+ in two steps. First the 3′ portion, codons 105 -185 (Figure 6) was inserted as a 330 bp NdeI - XbaI fragment. This fragment was generated by digesting 200 ng of the PCR product with NdeI and XbaI, and then was purified from an agarose gel. 50 ng of this fragment was ligated with 50 ng of NdeI - XbaI digested and gel purified pCWori+. The clone, pCW3′, was identified by hybridization using a probe made by random priming labelling (Sambrook et al, 1989, Molecular cloning: a laboratory manual), with DIG-dUTP, of 50 ng of the PCR product. The second step was to digest the PCR product with NdeI to generate a 318 bp fragment which encodes codons 1 - 104. After gel electrophoresis and

purification, 50 ng of this fragment was ligated to 50 ng of NdeI digested pCW3'. The clones containing a functional xylanase gene were identified by screening transformants on plates containing remazo-brilliant blue xylan (RBBX). Those colonies which made halos, were expressing th gene for xylanase (Kluepfel, 1988, Methods in Enzymology, 154:367-382, which is incorporated herein by reference). This clone was called pCWBSX::S100C.

A version of the *B. circulans* xylanase gene was synthesized to aid in subsequent mutagenesis studies, using the first 103 codons of the natural gene, and 82 codons derived from synthetic DNA fragments (pCWBCX3'SYN, Figure 7). The synthetic gene portion is identical to the natural gene except codons were used, which reflect the frequency of usage for specific amino acid residues in the genes of *E. coli.* The 103 codon portion was replaced with the equivalent portion from pCWBSX::S100C, by isolating the NheI-NdeI restriction fragment of pCWBSX::S100C and ligating it to NheI-NdeI digested pCWBCX3'SYN. After ligation, the mixture was transformed into *E. coli* MV1190. Clones were analyzed by DNA sequencing to verify they carried the S100C mutation. The resultant plasmid was called pCWBCX::S100C.

The plasmid pCWBCX::S100C was purified and digested with the restriction enzymes EagI and NsiI to remove the portion encoding codons 136 to 153. The large plasmid fragment was isolated from an agarose gel as described above. A set of synthetic oligonucleotides: Xy146C TOP (SEQ ID NO:7); Xy147C BOTTOM (SEQ ID NO:8) and Xy148C BOTTOM (SEQ ID NO:9) (as shown below) was synthesized to replace the EagI to NsiI fragment these oligonucleotides encoded the mutations at F146C, T147C, and N148C.

```
EagI                                                    NsiI
 137 138 139 140 141 142 143 144 145 146 147 148 149 150 151 152
GG CCG ACT GGT TCG AAC GCC ACC ATC ACT TGC ACT AAC CAT GTC AAT GCA Xy146C TOP
   C TGA CCA AGC TTG CGG TGG TAG TGA AAG ACA TTG GTA CAG TT      Xy147C BOTTOM
   C TGA CCA AGC TTG CGG TGG TAG TGA AAG TGA ACG GTA CAG TT      Xy148C BOTTOM
```

For the construction of restriction fragments for cassette mutagenesis, after kinase treatment, complementary oligonucleotides were mixed together (20 pmol of each oligonucleotide), and the mixture was heated to 80°C for 10 min. The mixture was then allowed to slowly cool to room temperature. Portions from the annealed oligonucleotide mixture were then used directly in ligation reactions with the previously digested and purified pCWBCX::S100C. After ligation and transformation, clones carrying the modified EagI-NsiI fragment were identified by hybridization with a DIG-ddUTP labelled oligonucleotide. Once hybridization positive colonies were identified, plasmid DNA was isolated and *E. coli* was re-transformed, to ensure the purity of the clone. DNA sequencing was performed to verify changes at the desired codons.

EXAMPLE 2

CONSTRUCTION OF THE MUTANT TS2

The plasmid pCWBCX3'SYN was used to transform *E. coli* RZ1032, and single stranded UDNA was prepared as previously described in Example 1. Two mutagenic oligonucleotides for making V98C, and A152C (see below) were used in an *in vitro* DNA synthesis reaction as described in Example 1.

SEQ ID NO:10

Codons 101 100 99 98 97 96 95 94

pCW V98C 5′ C ATC ACT TTT GCA AGT ACC TTT ATA 3′

SEQ ID NO:11

Codons 155 154 153 152 151 150 149

pCW A152C 5′ GGA TTT CCA GCA ATT CAC GTG 3′

After transformation, colonies were screened for hybridization to both mutagenic oligonucleotides. The xylanase gene from a hybridization positive clone, TS2, was completely sequenced to ensure only the desired mutations were present.

EXAMPLE 3

CONSTRUCTION OF THE MUTANT TS4

An additional thermostable mutant containing an inter-molecular disulfide bridge was constructed by a mutation at amino acid position 179 to change the amino acid from a serine to a cysteine (S179C). This mutant was constructed using cassette mutagenesis to make TS4a using the oligonucleotides: S179C-1 (SEQ ID NO:12); S179C-2 (SEQ ID NO:13) and S179C-3 (SEQ ID NO:14) shown below:

Codons 178 179 180 181 182 183 184 185

S179C-1 5′ CT GGA TGC TCC AAT GTG ACA GTG TGG TAA AGA TCT TGA

S179C-2 3′ TCG AGA CCT ACG AGG TTA CAC

S179C-3 3′ TGT CAC ACC ATT TCT AGA ACT

The plasmid pCWBCX3′SYN was digested with SacI and HindIII. The large fragment was purified from an agarose gel. The oligonucleotides for S179C were processed as described in Example 1, for cassette mutagenesis. This protein contained a single active thiol, however the protein dimerizes spontaneously. It forms a mixture based on the amount of free sulfhydryl from Ellmans reagent testing, and from purification of both the monomer and dim r from the mixture. This mutation was then combined

with TS1, by standard subcloning to produce a mutant called TS4. The mutant TS4M is the combined TS1 + S179C monomeric species and the TS4D is the dimeric species of this mutant. The purity of the monomeric and dimeric species used in the thermostability assays is shown in Figure 8.

EXAMPLE 4

CONSTRUCTION OF MUTATIONS AT THE N-TERMINUS OF *B. CIRCULANS* XYLANASE

Several mutant xylanases with mutations in the N-terminal region, have been constructed via the method of cassette mutagenesis. These mutants are either single, double or triple amino acid changes at positions 3, 4 or 8. All of the gene constructions involved ligations of 2 pairs of oligonucleotides which contained the mutations, into the NheI/BspEI linearized plasmid pXYbc (Figure 9), via the well established recombinant procedures of (i) phosphorylation of the oligonucleotides, (ii) their ligation with the linearized plasmid, (iii) transformation into the *E. coli* competent cells, (iv) identification of the mutant transformants via hybridization with the labelled oligonucleotide as probe, and (v) confirmation of the mutation through gene sequencing. These procedures have been fully described in the preceding Examples. The oligonucleotides used for the production of various N-terminus mutants are shown below.

(1) TS5a mutant, where the asparagine-8 (N-8) has been converted into tyrosine (Y).

The oligonucleotides for the cassette mutagenesis are XY8Y-1 (SEQ ID NO:15), XY8Y-2 (SEQ ID NO:16), XY-11-2

(SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18), and are as show below:

```
                                XY8Y-1                                  XY-11-2
      1  2  3  4  5  6  7  8  9 | 10 11 12 13 14 15 16 17 18 19 20 21 22 23
N-ter A  S  T  D  Y  H  Q  Y  H | T  D  G  G  G  I  V  N  A  V  H  G  S  G
5'-  CT AGT ACA GAT TAT TGG CAA TAT TGG|ACA GAC GGT GGC GGT ATC  GTT AAT GCC GTG AAC GGC T
      ____A TGT CTA ATA ACC GTT ATA ACC TGT CTG CCA CCG|CCA TAG CAA TTA CGG CAC TTG CCG AGG CC
      NheI                      XY8Y-2                XY-16-1                   BspEI
```

(2) TS6a mutant, where threonine-3 (T-3), aspartic acid-4 (D-4) and asparagine-8 (N-8) have been converted to glycine (G), tyrosine (Y) and tyrosine (Y), respectively.

The oligonucleotides for cassette mutagenesis are XY3G4, 8Y-1; (SEQ ID NO:19) XY3G4, 8Y-2 (SEQ ID NO:20); XY-11-2 and XY-16-1 wherein the first two oligonucleotides are shown below:

```
                    XY3G4,8Y-1
      1  2  3  4  5  6  7  8  9 10 11 12 13
N-ter A  S  G  Y  Y  H  Q  Y  H  T  D  G  G
 5'- CT AGT GGA TAC TAT TGG CAA TAT TGG
      ____A CCT ATG ATA ACC GTT ATA ACC TGT CTG CCA CCG
      NheI          XY3G4,8Y-2
```

(3) TS3a mutant was prepared from a pair of oligonucleotides with mixed bases at codons-4 and 8. The oligonucleotides for cassette mutagenesis are XY(1-8)Mu-1 (SEQ ID NO:21), XY(1-8)Mu-2 (SEQ ID NO:22), XY-11-2 and XY-

16-1 wherein the first two oligonucleotides are shown below:

XY(1-8)Mu-1

	1	2	3	4	5	6	7	8	9	10	11	12	13
N-ter	A	S	T	D/Y	Y	W	Q		W	T	D	G	G
5'-	CT	AGT	ACA	TAC	TAC	TGG	CAA	Tk1	TGG				
		A	TGT	CTG	ATG	ACC	GTT	Am1	ACC	TGT	CTG	CCA	CCG

NheI XY(1-8)Mu-2

k,l,m are mixed bases used in synthesis of oligonucleotide.

k = T+A+G

l = C+G

m = T+A+C

4) TS3 mutant was constructed by combining the mutations from TS1 with that from TS3a. The plasmid pXYbc was digested with Ssp I, Hind III and Avr II. Two fragments from this digest were isolated, a 2 KB Ssp I - Hind III (the vector portion of pXYbc) and a 245 bp Ssp I (a small portion of the vector and the first 78 codons of TS3a). A second plasmid pCW::TS1 was digested with Ssp I and Hind III and a 345 bp fragment was isolated (codons 79 - 185 of TS1). These three fragments were mixed together and ligated. After transformation, clones were picked from RBBX-agar plates if they produced clearing zones (xylanase activity). A clone was analyzed by DNA sequencing and was found to contain the desired mutations. This clone was designated TS3.

CHARACTERIZATION OF THE PROTEIN PRODUCTS

Protein purification

Protein samples were prepared from cells by first making an extract of the cells by grinding 10 g of the cell paste with 25 g of alumina powder. After grinding to a smooth mixture, small amounts (5 ml) of ice cold buffer A

(10mM sodium acetate, pH 5.5) were added and the mixture ground vigorously between additions. DNase, 10 μg/ml, was added to lower the viscosity of the extract. The alumina and cell debris were removed by centrifugation of the mixture at 8000 x g for 30 min. The supernatant (25 ml) was then dialyzed overnight at 4°C against 3 L buffer A, using dialysis tubing with a 3500 molecular weight cutoff. A slight precipitate formed in the dialysis bag, which is removed by centrifugation at 8000 x g for 15 min.

The cell extract was then pumped onto a 50 ml bed volume, S-sepharose fast flow, cation exchange column (Kabi-Pharmacia, Canada), equilibrated in buffer A. The xylanase was eluted with a 300 ml linear gradient of 0 to 0.3 M NaCl in buffer A at a flow rate of 3 ml/min. The xylanase elutes at 100 to 150 ml of the gradient. The fraction are checked on SDS-PAGE, and those fractions having most of the xylanase were pooled, and concentrated by ultrafiltration using 3000 dalton molecular weight cutoff membranes (Amicon YM3). The concentrated material, 5 ml, was then applied to a 1.5 cm x 85 cm TSK-HW50S gel filtration column, equilibrated in 50 mM ammonium acetate pH 6. The xylanase eluted at a volume of 90 to 100 ml. These fractions were analyzed by SDS-PAGE, and the peaks pooled as pure xylanase. The protein was quantified using the extinction co-efficient at 280 nm. ε 0.1% solution = 4.08. Typical purified yield from 10 g of cells was 25 mg of xylanase.

Detection of free thiol groups

Two methods were used to indicate the presence of the SS bond. The first was shown in Figure 10. Th electrophoretic mobility of the mutant proteins is faster under non-reducing conditions since the reduced protein is fully denatured and binds more SDS. This has also been observed for other disulfide bond containing proteins

(Mitchison and Wells, 1989, Biochemistry 28:4807-4815; Eder and Wilmanns, 1992, Biochemistry 31:4437-4444).

The second method checked for reactive sulfhydryl groups (SH groups). Approximately 200 μg of protein (10 nmol) were used for each determination. The reaction was performed in cuvettes at room temperature. The reaction mixture contained 6M urea, 50 mM Tris-HCl, pH 8, 1 mM EDTA, and 9 - 13 nmol of xylanase, in a final volume of 1 ml. The reaction was initiated by the addition of 10 μl of 10 mM 5,5'-dithiobis(2-nitrobenzoic acid), Ellmans reagent. The A_{412} of the solution was monitored for 45 min. The amount of free thiol was calculated from the liberated 2-nitro-5-thiobenzoate anion, using a molar extinction coefficient of 13,700. A single thiol containing mutant (S100C) was the positive control.

Titration of these and other mutants with Ellman's reagent under denaturing conditions shows undetectable levels of SH groups in the SS bond mutants, but shows stoichiometric amounts in a control containing a single SH group (Table 3). This shows that two of the mutants of the present invention (TS1 and TS2) have their cysteine residues in SS bridges.

Table 3.

Determination of free sulfhydryl groups in xylanase mutants

PROTEIN	[PROTEIN] μM (A_{280})	[SULFHYDRYL] μM	[SH]/ [PROTEIN]
BCX wild type	9.4	0	0
S100C	9.55	10.6	1.10
TS1	10.4	0	0
TS2	11.5	0	0
Mixture of S179C monomer and dimer	13.3	3	0.23

Measurement of enzymatic activity

The activity of the enzyme was measured two ways. The quantitative assay determined the number of reducing sugar ends generated from soluble xylan. The substrate for this assay was the fraction of birchwood xylan which dissolved in water from a 5% suspension of birchwood xylan (Sigma Chemical Co). After removing the insoluble fraction, the supernatant was freeze dried and stored in a desiccator. The measurement of specific activity was performed as follows. Reaction mixtures containing 100 μl of 30 mg/ml xylan in assay buffer (50 mM sodium citrate pH 5.5), 150 μl assay buffer, 50 μl of enzyme diluted in 1 mg/ml BSA, in assay buffer. The substrate and buffer were mixed and prewarmed at 40°C. The reaction was started by the addition of the enzyme. At various time intervals 50 μl portions were removed and the reaction stopped by diluting in 1 ml of 5 mM NaOH. The amount of reducing sugars was determined with the hydroxybenzoic acid hydrazide reagent (HBAH) (Lever, 1972, Analytical Biochem 47:273-279). A unit of enzyme activity was defined as that

amount generating 1 μmol reducing sugar in 1 minute at 40°C. For the determination of kinetic parameters substrate concentrations from 0.4 mg/ml to 20 mg/ml were used. Kinetic parameters were calculated using the computer program Enzfitter (Leatherbarrow 1987, Enzfitter, a non-linear regression data analysis program for the IBM-PC. Elsevier Science Publishers BV. Amsterdam, The Netherlands. 1987).

The second assay for activity was used for relative measurement, i.e. residual activity after heat treatment. This assay was performed with RBBX (Remazol Brilliant Blue Xylan) as the substrate (Biely et al, 1987, Methods in Enzymology 160: 536-541). 100 μl of 10 mg/ml RBBX in H_2O was mixed with 100 μl of 100 mM ammonium acetate pH 6, and the mixture prewarmed to 40°C. The reaction was started by the addition of suitably diluted enzyme. After a fixed length of time, 5 or 10 min, the reaction was stopped by the addition of 0.5 ml 95% ethanol. The mixture was inverted to mix, and then allowed to stand at least 10 min. at room temperature. The mixture was then centrifuged in a microcentrifuge for 3 min. at 12,000 X g. The A_{595} of the supernatant was measured.

Thermostability assay

To determine the thermostability of the mutant and wild type enzyme, the following parameters were used. The proteins were diluted to between 100 to 150 μg/ml, in dilution buffer (50 mM ammonium acetate pH 6.0). 400 μl of this solution were incubated in a 1.5 ml microcentrifuge tube, in a heating block containing glycerol. Portions of the solution were removed at specified times and immediately diluted 1 to 20 in dilution buffer and kept at room temperature until all samples had been taken. Portions of these diluted samples were then assayed for residual enzyme activity, using the RBBX assay. Results were expressed as percent residual activity compared to the

zero time point sample. Heating blocks were calibrated with a thermocouple and the measured temperature was that of the enzyme solution.

The data shown in Figures 11-16, clearly indicate the thermostabilizing effect of the introduction of non-native disulfide bridges into BCX. The mutant TS1 is clearly stable at 61°C for up to 3 h. The second SS bond mutant TS2 is not quite as stable, but maintains som activity after 1 h at 61°C (see Figure 12). Figures 11 and 12 show curves for the mixed TS4 monomer/dimer preparation, which is more stable than wild type but less stable than TS1. It is however, more relevant to examine the individual components. The mutant which contains only the inter-molecular SS bond (TS4a) shows increased thermostability at 58°C (Figure 13), and at this temperature is as stable as the intra-molecular SS bond mutant TS1 (Figure 14).

TS4 protein is produced as a mixture, but the monomer (TS4M) and the dimer (TS4D) are easily separated by rechromatographing the mixture on a cation exchange column. TS4M behaves very much like the TS1 mutant, whereas TS4D results in additive thermostability such that TS4D is more stable than TS1. (Figures 14, 15 and 16).

There has been another example of an artificial inter-molecular dimer using an SS bond, where the thermostability was increased, however this protein was not an enzyme, and the measurement of thermostability did not include a biological activity assay (Sauer et al, 1986, Biochemistry 25:5992-5998). The increase in thermostability seen in the intra-molecular SS bond mutants is similar to the effects seen in other proteins with an engineered intra-molecular SS bond; however, there is no significant decrease in specific activity, or change in kinetic parameters (Table 4), as is sometimes the case with

SS bond mutants (Kanaya et al, 1991, Journal of Biological Chemistry 266(10):6038-6044).

Table 4

Kinetic Parameters of Thermostable Xylanase Mutants

MUTANT	K_m (mg/ml)	V_{max} (μmol/min/mg)	k_{cat}/K_m
Wild type BCX	1.78±0.08	364±24	11.04
Wild type BSX	1.86±0.07	386±14	10.16
TS1	1.72±0.04	353±5	10.07
Wild type BCX°	2.21±0.27	456±46	10.086
TS2°	2.14±0.24	413±20	9.359
TS3°	2.27±0.22	269±1.4	5.819

° These values were determined with a different batch of substrate, than the first 3 were, which accounts for the difference in K_m and V_{max}

The decrease in the k_{cat}/K_m for TS3 indicates that some effect on catalysis and/or substrate specificity has occurred (Table 4). There is also a decrease in specific activity seen in TS4a and TS4D but this is not likely a severe limitation because decrease in the ability to hydrolyze soluble xylan may not be indicative of how well the protein performs in pre-bleaching applications on pulp (Table 5). The activity of the thermostable mutants is higher at elevated temperatures (Figure 17).

Table 5
Specific Activity of Thermostable Mutants of the *B. circulans* Xylanase

MUTATION	SPECIFIC ACTIVITY° (% OF WILD TYPE)
TS1	105
TS2	86
TS3a	77
TS3	61
TS4a	59
TS4M	97
TS4D	66
TS5a	93
TS6a	103

°These activities were determined using the reducing sugar assay (HBAH), and are the average of at least two determinations.

The N-terminal mutations, were found to confer thermostability, although less than that seen with the SS bond mutants. Thermostability of these mutants was determined in two ways: by a residual activity assay and by scanning calorimetry.

In the residual activity assay identical samples of a xylanase mutant were preheated in the assay buffer (50mM sodium citrate, pH 6.2) in Eppendorf tubes at different temperatures. The Eppendorf tubes with samples were heated in small water baths with lids. The heating temperatures were determined by a digital thermometer with a thermocouple sensor which measured to 0.1°C. After 30 min., samples were cooled down to 20°C before being assayed at 40°C. The residual enzyme activity of the heated sample

was expressed as a percentage of the activity of an unheated sample. This residual activity was plotted against the heating temperature (Figure 18). From the plot, the $T_{½}$ the preheating temperature, at which the xylanase still retained 50% of its activity, was determined and is shown in Table 6.

TABLE 6

$T_{1/2}$ of pre-heating temperatures at which xylanase will retain 50% of activity

Xylanase	$T_{½}$ (°C)
BCX (wild type)	53.5
TS3a	56.5
TS5a	55.5
TS6a	58.5

Samples of the xylanase were studied by scanning calorimetry to study the unfolding process of the xylanase structure as it relates to the rising temperature. Then T_m, the temperature at which half of the xylanase molecules have been denatured or melted, was determined. A higher T_m reflects a more thermostable molecule. It should be emphasized that calorimetry is concerned with the maintenance of the molecular structure, not enzymatic activity. However, the loss of enzyme activity through heating is often associated with the collapse of the molecular structure. The melting temperatures, T_m of wild type and mutant xylanases were determined from the calorimetric scans and are shown in Table 7.

TABLE 7

Melting temperature T_m of xylanas

Xylanase	T_m (°C)
BCX (wild type)	59.7
TS3a	62.7
TS5a	62.0
TS6a	64.6

Residue-8 appears to be essential to the thermostability of the BCX. The mechanism of the effect of this mutation may be explained by the 3-dimensional structure which reveals that a space existed between asparagine-8 and the other residues (Figure 19). This space was occupied by two water molecules in the wild-type enzyme (BCX). Molecular modelling indicated that substitution of asparagine-8 by phenylalanine or tyrosine could displace the water molecules (Figure 20). Increased thermostability resulting from this substitution may be a result of increased hydrophobic interaction contributed by these larger hydrophobic side-chains. The aromatic ring may displace the buried water molecules thus gaining hydrophobic interaction. In addition, the phenolic hydroxy group may also form a hydrogen bond with the carbonyl of peptide linkage-16 on the main chain.

The mutations at the N-terminus, described above, in combination with one of the SS bond mutations, resulted in an additive thermostability (Figure 14, 15 and 16). The combination of mutations in TS3 result in a protein with a thermostability of 64°C.

X-ray Crystallographic Structure of th Xylanases from *Bacillus circulans* and *Trichoderma harzianum*

Structure of the *B. circulans* xylanase

Crystals of the *B. circulans* xylanase were grown by the hanging drop vapour diffusion method. The reservoir buffer was 40mM Tris, pH 7.5, 22% saturated $(NH_4)_2SO_4$ and 100mM NaCl. Droplets were seeded after one day of equilibration. The space group of the crystals was $P2_12_12_1$. The unit cell parameters for all structures are given in Table 8. The heavy-atom derivative was obtained by soaking crystals of the Ser100Cys mutant protein in 10mM $HgCl_2$ for 6 days. All of the X-ray diffraction data sets for both the *B. circulans* and *T. harzianum* xylanase crystals were collected on a San Diego Multiwire Area Detector system on a Rigaku rotating anode generator.

All data reduction was performed using the San Diego software and the PHASES program package (Furey, W. & Swaminathan, S. (1990) "PHASES - A Program Package for the Processing and Analysis of Diffraction Data from Macromolecules", PA33, American Crystallographic Association Meeting Abstracts, Series 2, 18, pg 73). The initial electron density map for the *B. circulans* xylanase was calculated from phases based on the native (wild-type) data set and the positions of the mercury atoms found from the Patterson maps for the data collected on the $HgCl_2$ derivative of the Ser100Cys mutant protein. The initial electron density map was "skeletonized" using BONES (Greer, J. (1985) Methods in Enzymology 115, 206) and the initial model was built with TOM/FRODO (Jones, T. (1978) J. Appl. Cryst. 11, 268) and with O (Jones, T.A., Bergdoll, M., & Kjeldgaard, M. (1990) O: A macromolecular modelling environment. In "Crystallographic and Modelling Methods in Molecular Design" (Bugg, C.E. & Ealick, S.E., Editors).

Springer-Verlag, New York). All refinements were carried out using the simulated annealing and minimization protocols of X-PLOR (Brunger, A.T. (1988) J. Mol. Biol. 203, 803).

Structure of TS1 mutant of the *B. circulans* xylanase

Crystals of the TS1 mutant were grown as for the wild-type enzyme and were isomorphous with the crystals of the wild-type enzyme. A model for the cysteine side-chains was built into a difference electron density map that was calculated with data measured from the TS1 crystals and with phases calculated from the refined model of the wild-type structure.

Structure of the *T. harzianum* xylanase

Crystals of the *T. harzianum* xylanase were grown by the hanging drop vapour diffusion method. The reservoir buffer was 16mM Tris, pH 7.5, 20% saturated $(NH_4)_2SO_4$. Droplets were seeded after one day of equilibration. The space group of the crystals was also $P2_12_12_1$ but with different unit cell dimensions and with a different crystal packing arrangement than for the *B. circulans* crystals.

The structure was solved by standard molecular replacement methods with the programs MERLOT (Fitzgerald, P.M.D. (1988) J. Appl. Cryst. 21, 273-278) and BRUTE (Fujinaga, M. and Read, R. (1987) J. Appl. Crystallogr., 20, 517-521) using the *B. circulans* xylanase as the search model. Refinement statistics for all structures are given in Table 9 and plots of average main-chain B-factors for the three structures are shown in Figures 21, 22 and 23. Ramachandran diagrams indicating the stereochemical quality of the structures are shown in Figures 24 and 25.

Table 8

Data collection statistics

Data Set	*B. circulans*			*T. harzianum*
	Wild-type	S100C mutant + $HgCl_2$	TS1	Wild-type
Unit cell parameters (Å) a	44.00	43.96	43.90	43.67
b	52.78	52.71	52.88	94.65
c	78.39	78.41	78.43	51.55
Resolution (Å)	1.6	2.3	1.6	1.8
Merging R-factor (R_{sym})	0.028	0.062	0.062	0.111
Completeness of data set to resolution limit	94%	99%	99%	98%
Isomorphous difference (R_{iso})	n/a	0.098	0.077	n/a
Figure of merit	n/a	0.77	n/a	n/a

where,

$$R_{sym} = \frac{\Sigma |\langle I \rangle - I_{obs}|}{\Sigma |I_{obs}|}$$

<I> is the average intensity for a particular reflection and I_{obs} is one intensity measurement for that reflection.

$$R_{iso} = \frac{\Sigma |F_{deriv} - F_{nat}|}{\Sigma |F_{nat}|}$$

F_{deriv} and F_{nat} are the amplitudes of the structure factors for the derivative (S100C + $HgCl_2$ and TS1) and native (wild-type) data sets, respectively.

n/a = not applicable to this data set

Figure of merit is a measure of the quality of a derivative for calculating phases.

Table 9

Refinement statistics

Data Set	*B. circulans*		*T. harzianum*
	Wild-type	TS1	Wild-type
Number of non-hydrogen protein atoms	1448	1446	1473
Solvent molecules	141 waters + 1 sulfate	152 waters	59 waters
R-factor	0.163	0.172	0.208
RMS-deviations from ideality for			
bonds (Å):	0.009	0.009	0.010
angles (°):	1.691	1.783	1.318
dihedrals (°):	27.371	27.525	30.016
impropers (°):	1.312	1.351	2.326

where,

$$R\text{-}factor = \frac{\Sigma |F_{calc} - F_{obs}|}{\Sigma |F_{obs}|}$$

F_{calc} and F_{obs} are the amplitudes of the structure factors that are calculated from the refined model structure and from the measured reflections, resp ctively.

EXAMPLE 5

CONSTRUCTION OF OTHER MUTATIONS AT THE N-TERMINUS OF *B. CIRCULANS* XYLANASE

Additional N-terminal mutants have been constructed and tabulated (Table 10). Construction of mutants TS7a, TS8a, TS9a, TS10a, TS11a, TS12a, TS13a, TS14a, TS15a, TS16a, TS17 and TS19a has been accomplished via the identical protocol of cassette mutagenesis which previously yielded TS3a, TS5a and TS6a as described in Example 4 of the application. This involved ligation of various oligonucleotides to the NheI/BspEI linearized plasmid pXYbc (Figure 9). Another mutant TS18a was also prepared via the same protocol with the NheI/BamHI linearized plasmid pXYbc. Different overlapping oligonucleotides encoding various mutation have been synthesized.

(a) TS7a mutant, where asparagine-8 has been converted to phenylalanine. The oligonucleotides for cassette mutagenesis are XY8F-1 (SEQ ID NO:23), XY8F-2 (SEQ ID NO:24), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY8F-1 (SEQ ID NO:23)

1	2	3	4	5	6	7	8	9
A	S	T	D	Y	H	Q	F	H

5'-CT AGT ACA GAT TAT TGG CAA TTC TGG

A TGT CTA ATA ACC GTT AAG ACC TGT CTG CCA CCG

XY8F-2 (SEQ ID NO:24)

(b) TS8a mutant, where asparagine-8 has been converted to tryptophan. The oligonucleotides for cassette mutagenesis are XY8W-1 (SEQ ID NO:25), XY8W-2 (SEQ ID NO:26), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY8W-1 (SEQ ID NO:25)

	1	2	3	4	5	6	7	8	9
	A	S	T	D	Y	W	Q	W	W

5'-CT AGT ACA GAT TAT TGG CAA TGG TGG

A TGT CTA ATA ACC GTT ACC ACC TGT CTG CCA CCG

XY8W-2 (SEQ ID NO:26)

(c) TS9a mutant, where aspartic acid-4 has been converted to tyrosine. The oligonucleotides for cassette mutagenesis are XY4Y-1 (SEQ ID NO:27), XY4Y-2 (SEQ ID NO:28), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY4Y-1 (SEQ ID NO:27)

	1	2	3	4	5	6	7	8	9
	A	S	T	Y	Y	W	Q	N	W

5'-CT AGT ACA TAC TAC TGG CAG AAC TGG

A TGT ATG ATG ACC GTC AAG ACC TGT CTG CCA CCG

XY4Y-2 (SEQ ID NO:28)

(d) TS10a mutant, where aspartic acid-4 and asparagine-8 have both been converted to tyrosine. The oligonucleotides for cassette mutagenesis are XY4,8Y-1 (SEQ ID NO:29), XY4,8Y-2 (SEQ ID NO:30), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY4,8Y-1 (SEQ ID NO:29)

	1	2	3	4	5	6	7	8	9
	A	S	T	Y	Y	W	Q	Y	W

5'-CT AGC ACA TAC TAT TGG CAA TAT TGG

G TGT ATG ATA ACC GTT ATA ACC TGT CTG CCA CCG

XY4,8Y-2 (SEQ ID NO:30)

(e) TS11a mutant, where aspartic acid-4 and asparagine-8 have been converted to tyrosine and tryptophan respectively. The oligonucleotides for cassette mutagenesis are XY4Y8W-1 (SEQ ID NO:31), XY4Y8W-2 (SEQ ID NO:32), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY4Y8W-1 (SEQ ID NO:31)

1	2	3	4	5	6	7	8	9
A	S	T	Y	Y	W	Q	W	W

5'-CT AGC ACA TAC TAT TGG CAA TGG TGG

G TGT ATG ATA ACC GTT ACC ACC TGT CTG CCA CCG

XY4Y8W-2 (SEQ ID NO:32)

(f) TS12a mutant, where threonine-3 has been converted to glycine. The oligonucleotides for cassette mutagenesis are XY3G-1 (SEQ ID NO:33), XY3G-2 (SEQ ID NO:34), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY3G-1 (SEQ ID NO:33)

1	2	3	4	5	6	7	8	9
A	S	G	D	Y	W	Q	N	W

5'-CT AGC GGA GAC TAT TGG CAG AAT TGG

G CCT CTG ATA ACC GTT TTA ACC TGT CTG CCA CCG

XY3G-2 (SEQ ID NO:34)

(g) TS13a mutant, where threonine-3 and aspartic acid-4 have been converted to glycine and tyrosine respectively. The oligonucleotides for cassette mutagenesis are XY3G4Y-1 (SEQ ID NO:35), XY3G4Y-2 (SEQ ID NO:36), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY3G4Y-1 (SEQ ID NO:35)

1	2	3	4	5	6	7	8	9
A	S	G	Y	Y	W	Q	N	W

5'-CT AGC GGA TAC TAT TGG CAG AAT TGG

G CCT ATG ATA ACC GTT TTA ACC TGT CTG CCA CCG

XY3G4Y-2 (SEQ ID NO:36)

(h) TS14a mutant, where threonine-3, aspartic acid-4 and asparagine-8 have been converted to glycine, tyrosine and phenylalanine respectively. The oligonucleotides for cassette mutagenesis are XY3G4Y8F-2 (SEQ ID NO:37), XY3G4Y8F-3 (SEQ ID NO:38), XY-11-2 (SEQ ID NO:17) and XY-16-1 (SEQ ID NO:18).

XY3G4Y8F-2 (SEQ ID NO:37)

```
   1  2  3  4  5  6  7  8  9
   A  S  G  Y  Y  W  Q  F  W
5'-CT AGC GGA TAC TAT TGG CAA TTC TGG
      G CCT ATG ATA ACC GTT AAG ACC TGT CTG CCA CCG
```

XY3G4Y8F-3 (SEQ ID NO:38)

(i) TS15a mutant, where serine-22 has been converted to proline. The oligonucleotides for cassette mutagenesis are XY-11-3 (SEQ ID NO:39), XY-16-3 (SEQ ID NO:40), XY22P-1 (SEQ ID NO:41) and XY22P-2 (SEQ ID NO:42).

XY-11-3 (SEQ ID NO:39)

```
   1  2  3  4  5  6  7  8  9
   A  S  T  D  Y  W  Q  N  W
5'-CT AGC ACC GAT TAC TGG CAG AAC TGG
      G TGG CTA ATG ACC GTC TTG ACC TGT CTG CCA CCG
```

XY-16-3 (SEQ ID NO:40)

XY22P-1 (SEQ ID NO:41)

```
  10  11  12  13  14  15  16  17  18  19  20  21  22  23
   T   D   G   G   G   I   V   N   A   V   N   G   P   G
5'-ACA GAC GGT GGC GGT ATC GTT AAT GCC GTG AAC GGC C
                CCA TAG CAA TTA CGG CAC TTG CCG GGG CC
```

XY22P-2 (SEQ ID NO:42)

(j) TS16a mutant, where asparagine-8 and glycine-21 have been converted to phenylalanine and proline respectively. The oligonucleotides for cassette mutagenesis are XY8F-1 (SEQ ID NO:23), XY8F-2 (SEQ ID NO:24), XY21P-1 (SEQ ID NO:43) and XY21P-2 (SEQ ID NO:44).

XY21P-1 (SEQ ID NO:43)

```
  10  11  12  13  14  15  16  17  18  19  20  21  22  23
   T   D   G   G   G   I   V   N   A   V   N   P   S   G
5'-ACA GAC GGT GGC GGT ATC GTT AAT GCC GTG AAC CCA T
                CCA TAG CAA TTA CGG CAC TTG GGT AGG CC
```

XY21P-2 (SEQ ID NO:44)

(k) TS17a mutant, where asparagine-8 and serine-22 have been converted to phenylalanine and proline respectively. The oligonucleotides for cassette mutagenesis are XY8F-1 (SEQ ID NO:23), XY8F-2 (SEQ ID NO:24), XY22P-1 (SEQ ID NO:41) and XY22P-2 (SEQ ID NO:42).

(1) TS19a mutant, where threonine-3, aspartic acid-4, asparagine-8 and serine-22 have been converted to glycine, tyrosine, tyrosine and proline respectively. The oligonucleotides for cassette mutagenesis are XY3G4,8Y-1 (SEQ ID NO:19), XY3G4,8Y-2 (SEQ ID NO:20), XY22P-1 (SEQ ID NO:41) and XY22P-2 (SEQ ID NO:42).

(m) TS18a is a mutant where asparagine-8 and glycine-23 have been converted to phenylalanine and proline. The oligonucleotides for cassette mutagenesis in the NheI/BamHI linearized plasmid pXYbc are XY8F-1 (SEQ ID NO:23), XY8F-2 (SEQ ID NO:24), XY24P-1 (SEQ ID NO:45), XY23P-2 (SEQ ID NO:46), XY-12a (SEQ ID NO:47) and XY-15a (SEQ ID NO:48). The plasmid obtained was subcloned to yield TS18a.

XY24P-1 (SEQ ID NO:45)

```
   10  11  12  13  14  15  16  17  18  19  20  21  22  23  24
    T   D   G   G   G   I   V   N   A   V   N   G   S   G   P
5'-ACA GAC GGT GGC GGT ATC GTT AAT GCC GTG AAC GGC TCC GGA CCA
                CCA TAG CAA TTA CGG CAC TTG CCG AGG GGC CCT
```

XY23P-2 (SEQ ID NO:46)

```
  25  26  27  28  29  30  31  32  33  34  35  36  37  38  39
   N   Y   S   V   N   W   S   N   T   G   N   F   V   V   G
-AAT TAT AGC GTC AAT TGG TCT AAT ACT GGG AAC TTC GTA GTC GGA
-TTA ATA TCG CAG TTA ACC AGA TTA TGA CCC TTG AAG CAT CAG CCT
```

XY-12a (SEQ ID NO:47)

```
  40  41  42  43  44  45
   K   G   W   T   T   G
-AAA GGT TGG ACG ACA G
-TTT CCA ACC TGC TGT CCT AG
```

XY-15a (SEQ ID NO:48)

(n) TS20a mutant with extension from both the N-terminus (glycine-(-1)) and from the C-terminus (glycine-186 and cysteine-187), has conversion of alanine-1 into cysteine. Its construction was accomplished via a 2-step protocol identical to that described in Example 1 of the application. Initially this involved a polymerase chain reaction (PCR) with th template plasmid pXYbc (Figure 9)

and primers XY(-1)G1C (SEQ ID NO:49) and XY186G187C (SEQ ID NO:50). Subsequently the Nde/HindIII linearized PCR product was ligated into the identically linearized plasmid pCWBCX3'SYN.

XY(-1)G1C (SEQ ID NO:49)

NdeI -1 1 2 3 4 5 6

st G C S T D Y W

5'-CAT CGA TGC TTA GGA GGT CAT ATG GGA TGT AGC ACA GAT TAC TGG

7 8

Q N

-CAA AAC-3'

XY186G187C (SEQ ID NO:50)

179 180 181 182 183 184 185 186 187

S S N V T V W G C ter HindIII

3'-GA AGG TTG CAA TGT CAC ACC CCA ACG ATT TCT AGA ACT TCG AAT AGC

-TAC TAT TCG ACA-5'

(o) TS21a mutant which has extension from both the N-terminus (glycine-(-1)) and from the C-terminus (glycine-186 and cysteine-187), has conversion of alanine-1 and asparagine-8 into cysteine and phenylalanine respectively. Its construction was accomplished with the protocol described for TS20a, with differences in the PCR template (plasmid of TS7a; primers XY(-1)G1C8F (SEQ ID NO:51) and XY186G187C (SEQ ID NO:50)).

XY(-1)G1C8F (SEQ ID NO:51)

NdeI -1 1 2 3 4 5 6

st G C S T D Y W

5'-CAT CGA TGC TTA GGA GGT CAT ATG GGA TGT AGT ACA GAT TAT TGG

7 8

Q F

-CAA TTC-3'

Table 10

Xylanase N-terminal mutants

Name	Mutations	Type of Mutation
TS3a	D4Y N8F	N-terminal mutation
TS5a	N8Y	N-terminal mutation
TS6a	T3G D4Y N8Y	N-terminal mutation
TS7a	N8F	N-terminal mutation
TS8a	N8W	N-terminal mutation
TS9a	D4Y	N-terminal mutation
TS10a	D4Y N8Y	N-terminal mutation
TS11a	D4Y N8W	N-terminal mutation
TS12a	T3G	N-terminal mutation
TS13a	T3G D4Y	N-terminal mutation
TS14a	T3G D4Y N8F	N-terminal mutation
TS15a	S22P	N-terminal mutation
TS16a	N8F G21P	N-terminal mutation
TS17a	N8F S22P	N-terminal mutation
TS18a	N8F G23P	N-terminal mutation
TS19a	T3G D4Y N8Y S22P	N-terminal mutation
TS20a	(-1)G A1C 186G 187C	N/C-terminal SS bond
TS21a	N8F (-1)G A1C 186G 187C	N-terminal mutation N/C-terminal SS bond

Expression, extraction and purification

The expression, extraction, purification and assay processes for the mutant xylanases were identical to the protocols described in Example 4 of the application. The mutant xylanases have shown specific enzyme activity close (70% - 100%) to the wild type BCX.

Thermostability assay

Thermostability assay was identical to that described for the N-terminal mutant enzymes previously described in Example 4 of application, with a major difference in equipment used in the assay. Th water bath for incubating enzymes in Example 4 has a fluctuation of ±1.5°C, while the circulating water bath (Haake type F 4391) used in the present examples fluctuated at ±0.1°C. The new water bath should therefore yield more accurate data in subsequent assay. Enzyme was initially incubated in buffer (50mM sodium citrate, pH 5.5) without substrate (xylan) at a set temperature for 30 min. The residual enzymic activity was then determined at a lower temperature of 40°C via hydrolysis of soluble birchwood xylan, with the reducing sugar estimated by the hydroxybenzoic hydrazide reagent (HBAH). The thermostability of BCX and the earlier mutants TS1, TS2, TS3, TS3a, TS5a and TS6a has been reevaluated using the new equipment (Table 11).

The thermostability temperature $T_{1/2}$ at which various xylanase mutants retain 50% of activity was determined. The differences between the $T_{1/2}$ of mutants and wild type BCX have been tabulated (Table 11). The discrepancies in the $T_{1/2}$ values of the earlier mutants in Table 6 (of the parent application) and Table 11 were most likely due to the differences in performance of the two water baths. The newly established $T_{1/2}$ of BCX is 55°C, as compared to an earlier value of 53.5°C in Table 6.

Various mutations N8F (in TS7a, TS3a, TS14a), N8Y (in TS5a, TS6a, TS10a) and S22P (in TS15a, TS17a, TS19a), alone or in combination, caused a moderate increase in $T_{1/2}$ of 2-4°C, as compared to the greater gain of 5-8°C by the internal or N/C-terminal disulfide bond (in TS1, TS2, TS3, TS20a).

Temperature optimal and pH optimum in soluble xylan

The temperatur optimum assay was carried out substantially as for thermostability determinations except instead of incubating the enzymes in buffer without xylan for 30 min., xylanase is incubated in the same manner with xylan in the buffer. The modified process is designed to evaluate the hydrolysis of soluble xylan by the mutant enzyme at different temperatures for 30 min. The reducing sugar released was then determined by the HBAH reagent.

Higher temperature exerted dramatically differ nt effects on the hydrolysis of soluble xylan by wild type BCX and mutants (Figure 26). The temperature optimum T_{op} is defined as the highest temperature at which 150% relative enzymic activity was maintained as compared to performance at 40°. The temperature optimum T_{op} of BCX is 60°C. The change of temperature optimum of mutants versus wild type BCX was tabulated (Table 11).

Mutations such as N8Y (in TS5a, TS10a), N8F (in TS7a, TS3a), S22P (TS15a, TS17a), with only minor gain of 2-3°C in the thermostability assay, caused an elevation of T_{op} by 5°C (Table 11). The positive effect of mutations N8Y and N8F indicates that the aromatic residues tyrosine and phenylalanine are advantageous to the T_{op}. However, the mutation to another aromatic residue tryptophan (N8W in mutants TS8a and TS11a) lowered the T_{op} by 4-5°C. While the mutation S22P together with N8F elevated the T_{op} by 10°C in mutant TS17a, a similar conversion of the neighbouring residues to proline (G21P and G23P) dramatically lowered the T_{op} of TS16a and TS18a by as much as 11°C.

In contrast, the internal disulfide (in TS1, TS2), with a gain of 5-8°C in thermostability assay, demonstrated no gain in T_{op} (Table 11). The N/C-terminal

disulfide bond (in TS20a) with good thermostability of 5°C has only a moderate elevation of 3°C in T_{op} (Table 11).

The mutations T3G (in TS12a) or D4Y (in TS9a) independently caused a decrease of 3-9°C in T_{op} (Table 11). However, the combination of this pair of seemingly damaging mutations T3G and D4Y, with N8Y generated mutant TS6a which demonstrated a T_{op} increase of 4°C as compared to TS5a (N8Y). On the other hand, this conversion of mutant TS5a to TS6a resulted in a net loss of thermostability (1°C). Such opposite effect on thermostability and temperature optimal was also repeated in the conversion of mutant TS7a to TS14a. With a further addition of mutation S22P in mutant TS19a, an elevation of T_{op} as much as 14°C has been achieved. However, despite this gain in the T_{op}, the total gain of thermostability ($T_{1/2}$) remained not more than 4°C.

There is no clear correlation between the thermostable temperature ($T_{1/2}$) determined from incubation without substrate and the temperature optimum (T_{op}) in the assay with soluble xylan (Table 11). Therefore, a gain in thermostability does not necessarily correlate with a gain in temperature optimum to hydrolyse xylan or to bleach pulp.

The functional pH (pH optimum) of mutant enzymes was determined. Initially the enzyme was incubated in buffers of various pH without substrate at 50°C for 20 hr. Then the enzymic activity of the incubated enzyme was determined via the 4 min. 40°C assay conducted in the incubation buffer, instead of the 50mM sodium citrate (pH 5.5) normally used for such assay (Example 4). The pH optimum (pH_{op}) is defined as the highest pH at which the enzyme still maintains 50% of the maximal activity. Five mutants TS5a, TS7a, TS17a, TS19a and TS20a with elevated T_{op} were sel cted for this study. The enzymic activity of these mutants was compared to wild type BCX at different pH

(Figure 27). BCX can only maintain maximal activity up to pH 5.5 while the selected mutants can endure up to pH 7. Therefore the pH optimum of these mutants have been increased by more than 1 pH unit (Table 11).

Table 11

Changes of thermostability, temperature optimum and pH optimum of mutant xylanases in soluble xylan assay.

Name	Mutations	Type of Mutation	Buffer $\Delta T_{1/2}$ °C	Sol. xylan ΔT_{op} °C	Sol. xylan ΔpH_{op}
BCX	wild type		0	0	0
TS1	S100C N148C	internal SS bond	8	0	
TS2	V98C A152C	internal SS bond	5	0	
TS3	S100C N148C D4Y N8F	internal SS bond N-term. mutation	11	5	
TS3a	D4Y N8F	N-term. mutation	2	5	
TS5a	N8Y	N-term. mutation	3	5	1.2
TS6a	T3G D4Y N8Y	N-term. mutation	2	9	
TS7a	N8F	N-term. mutation	3	5	1.2
TS8a	N8W	N-term. mutation	-2	-5	
TS9a	D4Y	N-term. mutation		-3	
TS10a	D4Y N8Y	N-term. mutation	2	5	
TS11a	D4Y N8W	N-term. mutation	-3	-4	
TS12a	T3G	N-term. mutation		-9	
TS13a	T3G D4Y	N-term. mutation		0	
TS14a	T3G D4Y N8F	N-term. mutation	2	9	
TS15a	S22P	N-term. mutation		5	
TS16a	N8F G21P	N-term. mutation		-11	
TS17a	N8F S22P	N-term. mutation	4	10	1.1
TS18a	N8F G23P	N-term. mutation		-10	
TS19a	T3G D4Y N8Y S22P	N-term. mutation	4	14	1.1
TS20a	(-1)G A1C 186G 187C	N/C-term. SS bond	5	3	1.1
TS21a	N8F (-1)G A1C 186G 187C	N-term. mutation N/C-term. SS bond		8	

In summary, the appropriate mutations (S22P, N/C-terminal disulfide bond, N8F and N8Y alone or in combination with T3G and D4Y) can permit the mutant xylanase to hydrolyse xylan at higher temperature and pH.

Biobleaching of Pulp by Mutant Xylanases

Although the 30 min. soluble xylan assay determines the effectiveness of mutant xylanase in direct hydrolysis of xylan at high temperature, the usefulness of the enzyme as a bleaching agent can only be established via the pulp bleaching test. The protocol for such small scale test has been published in detail (Tolan and Canovas, 1992; Nissen et al, 1992, both incorporated herein by reference) and well-known to researcher in the field. Pulp suspension at different temperature or pH was treated with the enzyme for 2 hr. The bleaching effect was measured in brightness gain or whiteness of the pulp after bleaching. In this process, the temperature optimum (T_{op}) is defined as the highest temperature at which the enzyme still achieves a brightness gain of 0.4 in the bleached pulp. The pH optimum (pH_{op}) is defined as the highest pH at which the enzyme still achieved a brightness gain of 0.4 in the bleached pulp.

Several mutants were chosen for the pulp test at pH 8. While the wild type BCX functioned well at pH 7 with a temperature optimum of 54°C, it failed at pH 8 (Figure 28). In the same test at pH 8 (Table 12), mutants TS20a, TS17a and TS19a showed increase of T_{op} by 3, 10 and 14°C, respectively over BCX (at pH 7). Thus the mutant xylanase TS19a can efficiently bleach pulp at temperatures as high as 68°C (T_{op}). In addition to elevated temperature, the performance of these mutants at pH 8 also indicated an pH

upshift of 1 unit over BCX. Such gains in T_{op} and pH_{op} in pulp bleaching (Table 12) are in general agreement to the gains already determined through the soluble xylan assay (Table 11).

The mutant TS1, which has achieved greater thermostability ($T_{1/2}$) than the three mutants above (Table 11), was also tested for its effectiveness in pulp bleaching. However, its performance (temperature optimum of 54°C) was only equal to the wild type BCX (Table 12). This has already been predicted in the 30 min assay on soluble xylan (Table 11). The general lack of agreement between the thermostability ($T_{1/2}$) and the temperature optimal T_{op} in pulp bleaching (Table 12) or soluble xylan assay (Table 11) confirmed that greater thermostability may not be reflected in a better performance in pulp bleaching at elevated temperature.

In conclusion, appropriate mutations (N8Y, N8F, S22P and N/C- terminal disulfide bond; the combination of T3G, D4Y and N8Y or N8F) at the N-terminus region (1-25) region are beneficial in the performance of the enzyme in pulp bleaching or other applications at higher temperature and pH.

Table 12

Changes in Thermostability, Temperature Optimum and pH Optimum of mutant xylanases in soluble xylan and pulp bleaching.

Name	Mutations	Buffer $\Delta T_{1/2}$ °C	Soluble xylan ΔT_{op} °C	Soluble xylan ΔpH_{op}	Pulp bleaching ΔT_{op} °C	Pulp bleaching ΔpH_{op}
BCX	wild type	0	0	0	0	0
TS1	S100C N148C	8	0		0	
TS17a	N8F S22P	4	10	1.1	10	1
TS19a	T3G D4Y N8Y S22P	4	14	1.1	14	1
TS209a	(-1)G A1C 186G 187C	5	3	1.1	3	1

The invention has been described with reference to particular embodiments, although it is understood that the specific details shown are merely illustrative, and the invention may be carried out in other ways without departing from the spirit and the scope of the following claims.

THE EMBODIMENTS OF THE INVENTION IN WHICH AN EXCLUSIVE PROPERTY OR PRIVILEGE IS CLAIMED ARE DEFINED AS FOLLOWS:

1. A modified family G xylanas essentially having the structure of the *B. circulans* enzyme or mutated to essentially have this structure, wherein said xylanase has increased stability and wherein said xylanase is modified through either the introduction of at least one non-native disulfide bridge, the introductionn of at least one N-terminal mutation, or combinations thereof;

wherein the disulfide bridge is an intra-molecular bridge between a cysteine amino acid, which has been introduced on the last strand of beta-sheet III, and a cysteine amino acid, which has been introduced on the alpha helix, or on either side, adjacent to the alpha helix or the disulphide bridge is an inter-molecular bridge between two xylanase molecules, wherein a cysteine amino acid has been introduced in each of said two molecules, on the external region; and

wherein the N-terminal mutation is selected from at least one mutation of the group consisting of the introduction of tyrosine at amino acid position 8, introduction of phenylalanine at amino acid position 8, introduction of proline at amino acid position 22, and introduction of an N-terminal to C-terminal disulfide bridge, and wherein these N-terminal mutations can be used in combination with other N-terminal mutation introduced at amino acid position 1 to 25 of the N-terminal region, based on the amino acid numbering from *B. circulans* xylanase.

2. A modified family G xylanase of Claim 1, wherein said xylanase has increased stability as demonstrated by at least one of an increase in thermostability, an increase in temperature optimum, an increase in pH optimum, or combinations thereof, as determined in the hydrolysis of xylan or in the bleaching of pulp.

3. The modified xylanase of Claim 1, wherein the family G xylanase is select d from a bacterial or fungal xylanase.

4. The modified xylanase of Claim 3, wherein the bacterial xylanase is from bacteria selected from the group consisting of *Bacillus pumilus; Clostriduim acetobutylicum; Ruminococcus flavefaciens; Streptomyces sp.; Streptomyces lividans, B; Streptomyces lividans, C; Bacillus circulans; and Bacillus subtilis*.

5. The modified xylanase of Claim 3, wherein the fungal xylanase is from fungi selected from the group consisting of *Trichoderma reesei, II; Trichoderma viride; Trichoderma harzianum; Schizophyllum commune; Aspergillus niger var. awamori; Aspergillus nubigensis, A; and Trichoderma reesei, I.*

6. The modified xylanase of Claim 1, wherein the cysteine on the last strand of beta-sheet III is between amino acid residues 95 and 109 and the cysteine on the alpha-helix is between the amino acid residues from 143 to 158, based on the amino acid numbering from *B. circulans* xylanase.

7. The modified xylanase of Claim 1, wherein the disulfide bridge is an intra-molecular bridge between amino acids selected from the group consisting of 100 and 148, and 98 and 152, based on the amino acid numbering from *B. circulans* xylanase.

8. The modified xylanase of Claim 1, wherein the cysteine amino acid is introduced on an external region of the xylanase molecule selected from the regions consisting of b ta-sheet I, the loop preceding the last strand of sheet III and beta-sheet III, and alpha-helix and surrounding loops.

9. The modified xylanase of Claim 8, wh rein the cysteine amino acid is introduced at a residue position selected from the group consisting of amino acid 15 to 31 of beta-sheet I, amino acid 43 to 61 of beta-sheet I, amino acid 87 to 104 of the loop and beta-sheet III, amino acid 133 to 163 of the alpha-helix and surrounding loops, and amino acid 177 to 185 of beta-sheet I, based on the amino acid numbering from *B. circulans* xylanase.

10. The modified xylanase of Claim 9, wherein the cysteine amino acid is located at amino acid position 179, based on the amino acid numbering from *B. circulans* xylanase.

11. A modified family G xylanase of Claim 1, wher in the N-terminal to C-terminal disulfide bond results from an extension at the N-terminus and C-terminus end of the protein to add cysteine or by the conversion of amino acids at the termini to cysteine.

12. The modified family G xylanase of Claim 11, wherein the N-terminus is extended by adding a glycine at position -1, the C-terminus is extended by adding glycine and cysteine at position 186 and 187, respectively and wherein the alanine at position 187 is converted into cysteine.

13. The modified xylanase of Claim 1, wherein the xylanase is modified at the N-terminus by combining a mutation selected from the group consisting of: th introduction of tyrosine at amino acid position 8, the introduction of phenylalanine at amino acid position 8, the introduction of proline at amino acid position 22, and the introduction of an N-terminal to C-terminal disulfide bridge, with a mutation at position 3 or 4.

14. The modified xylanase of Claim 1, wherein said modified xylanase are produced from clones selected from the group consisting of TS1, TS2, TS3, TS3a, TS5a, TS6a, TS7a, TS10a, TS14a, TS15a, TS17a, TS19a, TS20a and TS21a.

15. The modified xylanase of Claim 14, wherein said modified xylanase are produced from clones selected from the group consisting of TS3, TS3a, TS5a, TS6a, TS7a, TS10a, TS14a, TS15a, TS17a, TS19a, TS20a and TS21a.

16. The modified xylanase of Claim 15, wherein said modified xylanase are produced from clones selected from the group consisting of TS6a, TS14a, TS17a, TS19a and TS21a.

17. A modified family G xylanase essentially having the structure of the *B. circulans* enzyme or mutated to essentially have this structure, wherein said xylanase has increased stability and wherein said xylanase is modified through either the introduction of at least one non-native disulfide bridge, the introduction of at least one N-terminal mutation, or combinations thereof; and wherein said modified xylanase is produced from clones selected from the group consisting of TS1, TS2, TS3, TS3a, TS5a, TS6a, TS7a, TS10a, TS14a, TS15a, TS17a, TS19a, TS20a and TS21a.

18. A modified family G xylanase essentially having the structure of the *B. circulans* enzyme or mutated to essentially have this structure, wherein said xylanase has increased stability and wherein said xylanase is modified through either the introduction of at least one non-native disulfide bridge, the introduction of at least one N-terminal mutation, or combinations thereof; and wherein said modified xylanase is produced from clones selected from the group consisting of TS6a, TS14a, TS17a, TS19a and TS21a.

```
BP       1   RTITNNEMGN HSGYDYELWK DYGNT-SMTL NNGGAFSAGW N--NIGNA   45
CA      32   KTITSNEIGV NGGYDYELWK DYGNT-SMTL KNGGAFSCQW S--NIGNA   76
RF       1   SAADQQTRGN VGGYDYEMWN QNGQGQASMN PGAGSFTCSW S--NIENF   46
TR2      1    QTIQPGTGY NNGYFYSYWN DGHGGVTYTN GPGGQFSVNW S--NSGNF   45
TV       1    QTIQPGTGF NNGYFYSYWN DGHGGVTYTN GPGGQFSVNW S--NSGNF   45
TH       1    QTIGPGTGY SNGYYYSYWN DGHAGVTYTN GGGGSFTVNW S--NSGNF   45
SC       1    SGTPSSTGT DGGYYYSWWT DGAGDATYQN NGGGSYTLTW SG-NNGNL   46
AN       1           S AGINYVQNYN GNLGDFTY-D ESAGTFSMYW EDGVSSDF   37
AT       1             AGINYVQNYN QNLGDFTY-D ESAGTFSMYW EDGVSSDF   37
TR1      1             ASINYDQNYQ TGG-QVSYS- PSNTGFSVNW N--TQDDF   34
SS       1 ATTIT-NETGY D-GMYYSFWT DGGGSVSMTL NGGGSYSTRW T--NCGNF   45
SLB      1 DTVVTTNQEGT NNGYYYSFWT DSQGTVSMNM GSGGQYSTSW R--NTGNF   47
SLC      1 ATTITTNQTGT D-GMYYSFWT DGGGSVSMTL NGGGSYSTQW T--NCGNF   46
BC       1             ASTDYWQNWT DGGGIVNAVN GSGGNYSVNW S--NTGNF   36
BS       1             ASTDYWQNWT DGGGIVNAVN GSGGNYSVNW S--NTGNF   36

Consensus  .........G. ..GYYY..W. DGGG.V.... ..GG.FS..W S..N.GNF

BP      46   LFRK-GKKFD ST-RTHHQLG NISINYNASF N-PSGNSYLC VYGWTQSP   90
CA      77   LFRK-GKKFN DT-QTYKQLG NISVNYDCNY Q-PYGNSYLC VYGWTSSP  121
RF      47   LARM-GKNYD SQKKNYKAFG NIVLTYDVEY T-PRGNSYMC VYGWTRNP   92
TR2     46   VGGK-GWQPG TKNKV----- ---INFS-GS YNPNGNSYLS VYGWSRNP   83
TV      46   VGGK-GWQPG TKNKV----- ---INFS-GS YNPNGNSYLS VYGWSRNP   83
TH      46   VGGK-GWQPG TKNKV----- ---INFS-GS YNPNGNSYLS IYGWSRNP   83
SC      47   VGGK-GWNPG AASRS----- ---ISYS-GT YQPNGNSYLS VYGWTRSS   84
AN      38   VVGL-GWTTG SSNA------ ---ITYSAEY SASGSSSYLA VYGWVNYP   76
AT      38   VVGLGGWTTG SSNA------ ---ITYSAEY SASGSASYLA VYGWVNYP   76
TR1     35   VVGV-GWTTG SSAP------ ---INFGGSF SVNSGTGLLS VYGWSTNP   72
SS      46   VAGK-GWANG GR-RT----- ---VRYT-GW FNPSGNGYGC LYGWTSNP   82
SLB     48   VAGK-GWANG GR-RT----- ---VQYS-GS FNPSGNAYLA LYGWTSNP   84
SLC     47   VAGK-GWSTG DGN------- ---VRYN-GY FNPVGNGYGC LYGWTSNP   82
BC      37   VVGK-GWTTG SPFRT----- ---INYNAGV WAPNGNGYLT LYGWTRSP   75
BS      37   VVGK-GWTTG SPFRT----- ---INYNAGV WAPNGNGYLT LYGWTRSP   75

Consensus  V.GK.GW..G .......... ...INY..G. ..P.GNSYL. VYGWT.NP

BP      91   LAEYYIVDSW GTYR-PT--G AYKGSFYADG GTYDIYETTR VNQPSIIG  135
CA     122   LVEYYIVDSW GSWRPP--GG TSKGTITVDG GIYDIYETTR INQPSIQG  167
RF      93   LMEYYIVEGW GDWRPPGNDG EVKGTVSANG NTYDIRKTMR YNQPSLDG  140
TR2     84   LIEYYIVENF GTYN-PSTGA TKLGEVTSDG SVYDIYRTQR VNQPSIIG  130
TV      84   LIEYYIVENF GTYN-PSTGA TKLGEVTSDG SVYDIYRTQR VNQPSIIG  130
TH      84   LIEYYIVENF GTYN-PSTGA TKLGEVTSDG SVYDIYRTQR VNQPSIIG  130
SC      85   LIEYYIVESY GSYD-PSSAA SHKGSVTCNG ATYDILSTWR YNAPSIDG  131
AN      77   GAEYYIVEDY GDYN-PCSSA TSLGTVYSDG STYQVCTDTR INEPSITG  123
AT      77   QAEYYIVEDY GDYN-PCSSA TSLGTVYSDG STYQVCTDTR INEPSITG  123
TR1     73   LVEYYIMEDN HNY--PAQ-G TVKGTVTSDG ATYTIWENTR VNEPSIQG  117
SS      83   LVEYYIVDNW GSYR-PT--G ETRGTVHSDG GTYDIYKTTR YNAPSVEA  127
SLB     85   LVEYYIVDNW GTYR-PT--G EYKGTVTSDG GTYDIYKTTR VNKPSVEG  129
SLC     83   LVEYYIVDNW GSYR-PT--G TYKGTVSSDG GTYDIYQTTR YNAPSVEG  127
BC      76   LIEYYVVDSW GTYR-PT--G TYKGTVKSDG GTYDIYTTTR YNAPSIDG  120
BS      76   LIEYYVVDSW GTYR-PT--G TYKGTVKSDG GTYDIYTTTR YNAPSIDG  120

Consensus  L.EYYIVE.W G.YR.P...G T.KGTV.SDG .TYDIY.TTR .N.PSI.G
```

FIGURE 1

```
BP    136   -IATFKQYWS VRQTKRTS-- ------GTVS VSAHFRKWES LGMPM-GK  174
CA    168   -NTTFKQYWS VRRTKRTS-- ------GTIS VSKHFAAWES KGMPL-GK  206
RF    141   -TATFPQYWS VRQTSGSANN QTNYMKGTID VSKHFDAWSA AGLDMSGT  187
TR2   131   -TATFYQYWS VRRNHR-S-S ------GSVN TANHFNAWAQ QGLTL-GT  169
TV    131   -TATFYQYWS VRRTHR-S-S ------GSVN TANHFNAWAQ QGLTL-GT  169
TH    131   -TATFYQYWS VRRNHR-S-S ------GSVN TANHFNAWAS HGLTL-GT  169
SC    132   -TQTFEQFWS VRNPKKAPGG SIS---GTVD VQCHFDAWKG LGMNLGSE  175
AN    124   -TSTFTQYFS VRESTRTS-- ------GTVT VANHFNFWAQ HGFGN-SD  162
AT    124   -TSTFTQYFS VRESTRTS-- ------GTVT VANHFNFWAH HGFHN-SD  162
TR1   118   -TATFNQYIS VRNSPR-T-S ------GTVT VQNHFN-WAS LGLHLGQM  155
SS    128   -PAAFDQYWS VRQSKVT--S ------GTIT TGNHFDAWAR AGMNMGNF  168
SLB   130   TR-TFDQYWS VRQSKR-TG- ------GTIT TGNHFDAWAR AGMPLGNF  168
SLC   128   TK-TFQQYWS VRQSKVTSGS ------GTIT TGNHFDAWAR AGMNMGQF  168
BC    121   DRTTFTQYWS VRQSKRPTGS N-----ATIT FTNHVNAWKS HGMNLGSN  163
BS    121   DRTTFTQYWS VRQSKRPTGS N-----ATIT FSNHVNAWKS HGMNLGSN  163

Consensus   .T.TF.QYWS VR.SKR.S.. ......GTVT ..NHFNAWA. .GH.L...


BP    175   MYETAFTVEG YQSSGSANVM TNQLFIGN                        201
CA    207   MHETAFNIEG YQSSGKADVN SMSINIGK                        233
RF    188   LYEVSLNIEG YRSNGSANVK SVSV                            211
TR2   170   MDYQIVAVEG YFSSGSASI- TVS                              190
TV    170   MDYQIVAVEG YFSSGSASI- TVS                              190
TH    170   MDYQIVAVEG YFSSGSASI- TVS                              190
SC    176   HNYQIVATEG YQSSGTATI- TVT                              197
AN    163   FNYQVMAVEA WSGAGSASV- TISS                             184
AT    163   FNYQVVAVEA WSGAGSAAV- TISS                             184
TR1   157   MNYQVVAVEG WGGSGSASQ- SVSN                             178
SS    167   RYYMIMATEG YQSSGSSTI- TVSG                             189
SLB   169   SYYMIMATEG YQSSGTSSI- NVGGTGGGDS GGATTGAVAA GAPTVSAG  215
SLC   169   RYYMIMATEG YQSSGSSNI- TVSG                             191
BC    164   WAYQVMATEG YQSSGSSNV- TVW                              185
BS    164   WAYQVMATEG YQSSGSSNV- TVW                              185

Consensus   ..YQ..A.EG YQSSGSA... TVS

**********************************************************************
```

BP *Bacillus pumilus*
CA *Clostridium acetobutylicum*, Xyn B
RF *Ruminococcus flavefaciens*
TR2 *Trichoderma reesei*, XYN II, 21kD, pI 9.0
TV *Trichoderma viride*, 20kD
TH *Trichoderma harzianum*, 20kD
SC *Schizophyllum commune*, Xylanase A
AN *Aspergillus niger*, var. awamori
AT *Aspergillus tubigensis*, A
TR1 *Trichoderma reesei*, XYN I, 19kD, pI 5.2
BS *Bacillus subtilis*
BC *Bacillus circulans*
SLB *Streptomyces lividans*, Xln B
SLC *Streptomyces lividans*, Xln C
SS *Streptomyces* sp. #36a

FIGURE 1 CONT'D

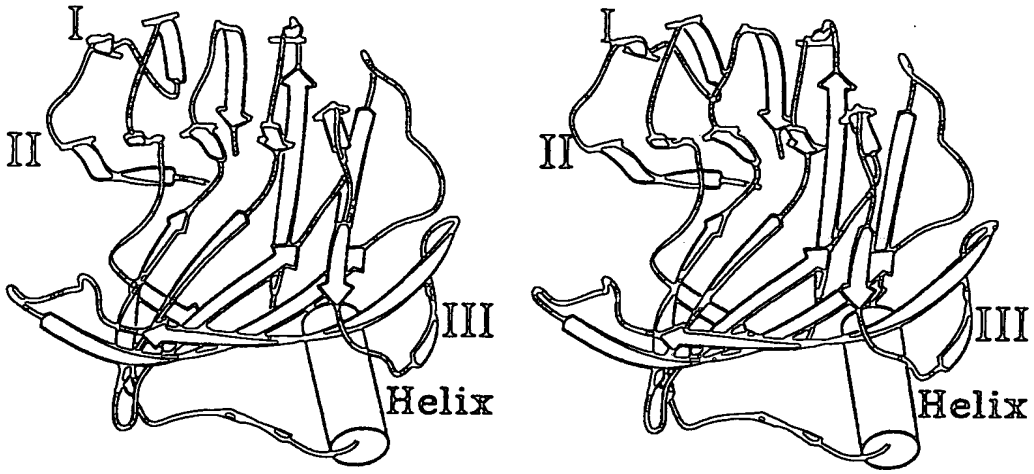


FIGURE 2

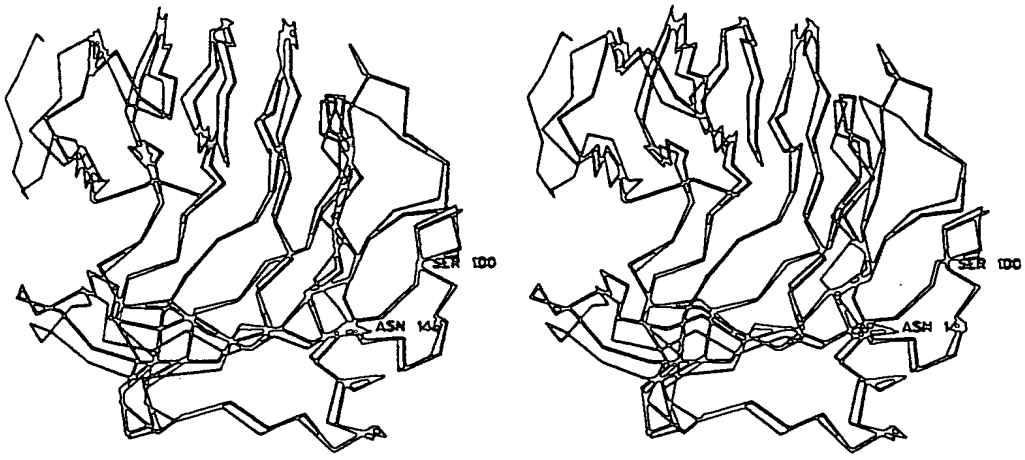


FIGURE 3

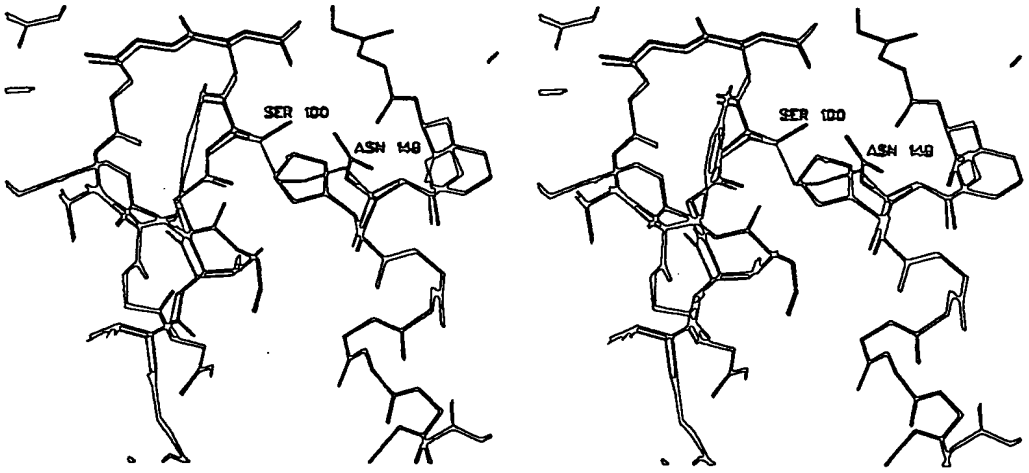


FIGURE 4

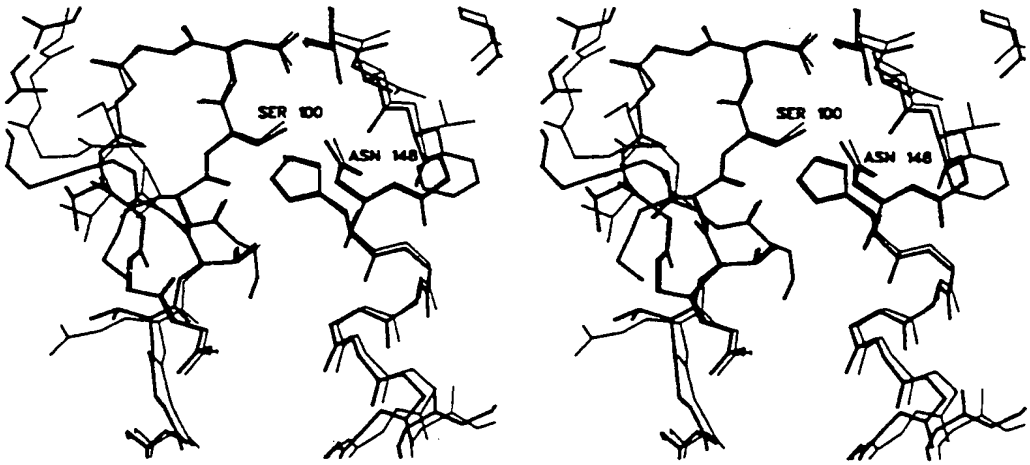


FIGURE 5

```
 1   2   3   4   5   6   7   8   9  10  11  12  13  14  15  16
 A   S   T   D   Y   W   Q   N   W   T   D   G   G   G   I   V
GCT AGC ACA GAC TAC TGG CAA AAT TGG ACT GAT GGG GGC GGT ATA GTA
CGA TCG TGT CTG ATG ACC GTT TTA ACC TGA CTA CCC CCG CCA TAT CAT
 NheI     5' PCR primer site

 17  18  19  20  21  22  23  24  25  26  27  28  29  30  31  32
 N   A   V   N   G   S   G   G   N   Y   S   V   N   W   S   N
AAC GCT GTC AAT GGG TCT GGC GGG AAT TAC AGT GTT AAT TGG TCT AAT
TTG CGA CAG TTA CCC AGA CCG CCC TTA ATG TCA CAA TTA ACC AGA TTA

 33  34  35  36  37  38  39  40  41  42  43  44  45  46  47  48
 T   G   N   F   V   V   G   K   G   W   T   T   G   S   P   F
ACC GGA AAT TTT GTT GTT GGT AAA GGT TGG ACT ACA GGT TCG CCA TTT
TGG CCT TTA AAA CAA CAA CCA TTT CCA ACC TGA TGT CCA AGC GGT AAA

 49  50  51  52  53  54  55  56  57  58  59  60  61  62  63  64
 R   T   I   N   Y   N   A   G   V   W   A   P   N   G   N   G
AGG ACG ATA AAC TAT AAT GCC GGA GTT TGG GCG CCG AAT GGC AAT GGA
TCC TGC TAT TTG ATA TTA CGG CCT CAA ACC CGC GGC TTA CCG TTA CCT
                                      NaeI

 65  66  67  68  69  70  71  72  73  74  75  76  77  78  79  80
 Y   L   T   L   Y   G   W   T   R   S   P   L   I   E   Y   Y
TAT TTA ACT TTA TAT GGT TGG ACG AGA TCA CCT CTC ATA GAA TAT TAT
ATA AAT TGA AAT ATA CCA ACC TGC TCT AGT GGA GAG TAT CTT ATA ATA
                                              SspI

 81  82  83  84  85  86  87  88  89  90  91  92  93  94  95  96
 V   V   D   S   W   G   T   Y   R   P   T   G   T   Y   K   G
GTA GTG GAT TCA TGG GGT ACT TAT AGA CCT ACT GGA ACG TAT AAA GGT
CAT CAC CTA AGT ACC CCA TGA ATA TCT GGA TGA CCT TGC ATA TTT CCA

 97  98  99 100 101 102 103 104 105 106 107 108 109 110 111 112
 T   V   K   S   D   G   G   T   Y   D   I   Y   T   T   T   R
ACT GTA AAA AGT GAT GGG GGT ACA TAT GAC ATA TAT ACA ACT ACA CGT
TGA CAT TTT TCA CTA CCC CCA TGT ATG CTG TAC ATA TGT TGC TGT GCA
S100C mutagenic primer site  NdeI

113 114 115 116 117 118 119 120 121 122 123 124 125 126 127 128
 Y   N   A   P   S   I   D   G   D   R   T   T   F   T   Q   Y
TAT AAC GCA CCT TCC ATT GAT GGC GAT CGC ACT ACT TTT ACG CAG TAC
ATA TTG CGT GGA AGG TAA CTA CCG CTA GCG TGA TGA AAA TGC GTC ATG
                                  PvuI

129 130 131 132 133 134 135 136 137 138 139 140 141 142 143 144
 W   S   V   R   Q   S   K   R   P   T   G   S   N   A   T   I
TGG AGT GTT CGC CAG TCG AAG AGA CCA ACC GGA AGC AAC GCT ACA ATC
ACC TCA CAA GCG GTC AGC TTC TCT GGT TGA CCA TCG TTG CGA TGT TAG
```

FIGURE 6

```
145 146 147 148 149 150 151 152 153 154 155 156 157 158 159 160
 T   F   S   N   H   V   N   A   W   K   S   H   G   M   N   L
ACT TTC AGC AAT CAT GTG AAC GCA TGG AAG AGC CAT GGA ATG AAT CTG
TGA AAG TCG TTA GTA CAC TTG CGT ACC TTC TCG GTA CCT TAC TTA GAT
                                          NcoI

161 162 163 164 165 166 167 168 169 170 171 172 173 174 175 176
 G   S   N   W   A   Y   Q   V   M   A   T   E   G   Y   Q   S
GGC AGT AAT TGG GCT TAC CAA GTC ATG GCG ACA GAA GGA TAT CAA AGT
CCG TCA TTA ACC CGA ATG GTT CAT TAC CGC TGT CTT CCT ATA GAT TCA
                                              EcoRV

177 178 179 180 181 182 183 184 185 STOP
 S   G   S   S   N   V   T   V   W
AGT GGA AGT TCT AAC GTA ACA GTG TGG TAA CAGATCATCC TTAATCAGGG
TCA CCT TCA AGA TTG CAT TGT CAC ACC ATT GTCTAGTAGG AATTAGTCCC

GTAGCTAACG GGCTGCTGAT CGTTCCTTGA GAAGTTTTA TAATCAATGA TTATTAAAAT
CATCGATTGC CCGACGACTA GCAAGGAACT CTTCAAAAT ATTAGTTACT AATAATTTTA
                                                    3' PCR primer

CGTTAGTAA TGGTTAAAGG TTGTTTTCTA CTAGGTGAAC GGCCTTGGAA TTGCTGGAGG
GCAATCATT ACCAATTTCC AACAAAAGAT GATCCACTTG CCGGAACCTT AACGACCTCC
binding site

TAGGGTATTC TCCATCTGGT TTTT
ATCCCATAAG AGGTAGACCA AAAA
```

FIGURE 6 CONT'D

```
 1   2   3   4   5   6   7   8   9  10  11  12  13  14  15  16
 A   S   T   D   Y   W   Q   N   W   T   D   G   G   G   I   V
GCT AGC ACA GAC TAC TGG CAA AAT TGG ACT GAT GGG GGC GGT ATA GTA
CGA TCG TGT CTG ATG ACC GTT TTA ACC TGA CTA CCC CCG CCA TAT CAT
 NheI

 17  18  19  20  21  22  23  24  25  26  27  28  29  30  31  32
 N   A   V   N   G   S   G   G   N   Y   S   V   N   W   S   N
AAC GCT GTC AAT GGG TCT GGC GGG AAT TAC AGT GTT AAT TGG TCT AAT
TTG CGA CAG TTA CCC AGA CCG CCC TTA ATG TCA CAA TTA ACC AGA TTA

 33  34  35  36  37  38  39  40  41  42  43  44  45  46  47  48
 T   G   N   F   V   V   G   K   G   W   T   T   G   S   P   F
ACC GGA AAT TTT GTT GTT GGT AAA GGT TGG ACT ACA GGT TCG CCA TTT
TGG CCT TTA AAA CAA CAA CCA TTT CCA ACC TGA TGT CCA AGC GGT AAA

 49  50  51  52  53  54  55  56  57  58  59  60  61  62  63  64
 R   T   I   N   Y   N   A   G   V   W   A   P   N   G   N   G
AGG ACG ATA AAC TAT AAT GCC GGA GTT TGG GCG CCG AAT GGC AAT GGA
TCC TGC TAT TTG ATA TTA CGG CCT CAA ACC CGC GGC TTA CCG TTA CCT
                                    NaeI

 65  66  67  68  69  70  71  72  73  74  75  76  77  78  79  80
 Y   L   T   L   Y   G   W   T   R   S   P   L   I   E   Y   Y
TAT TTA ACT TTA TAT GGT TGG ACG AGA TCA CCT CTC ATA GAA TAT TAT
ATA AAT TGA AAT ATA CCA ACC TGC TCT AGT GGA GAG TAT CTT ATA ATA
                                            SspI

 81  82  83  84  85  86  87  88  89  90  91  92  93  94  95  96
 V   V   D   S   W   G   T   Y   R   P   T   G   T   Y   K   G
GTA GTG GAT TCA TGG GGT ACT TAT AGA CCT ACT GGA ACG TAT AAA GGT
CAT CAC CTA AGT ACC CCA TGA ATA TCT GGA TGA CCT TGC ATA TTT CCA

 97  98  99 100 101 102 103 104 105 106 107 108 109 110 111 112
 T   V   K   S   D   G   G   T   Y   D   I   Y   T   T   T   R
ACT GTA AAA AGT GAT GGG GGT ACA TAT GAC ATC TAC ACC ACC ACA AGA
TGA CAT TTT TCA CTA CCC CCA TGT ATG CTG TAG ATG TGG TGG TGT TCT
                           NdeI
V908C mutagenic primer site

113 114 115 116 117 118 119 120 121 122 123 124 125 126 127 128
 Y   N   A   P   S   I   D   G   D   R   T   T   F   T   Q   Y
TAC AAC GCA CCT TCC ATC GAT GGC GAT CGG ACC ACC TTT ACT CAG TAC
ATG TTG CGT GGA AGG TAG CTA CCG CTA GCC TGG TGG AAA TGA GTC ATG
                    ClaI      PvuI

129 130 131 132 133 134 135 136 137 138 139 140 141 142 143 144
 W   S   V   R   Q   S   K   R   P   T   G   S   N   A   T   I
TGG AGT GTT AGA CAA TCT AAG CGG CCG ACT GGT TCG AAC GCC ACC ATT
ACC TCA CAA TCT GTT AGA TTC GCC GGG TGA CCA AGC TTG CGG TGG TAA
                          EagI/EaeI     BstBI
```

FIGURE 7

```
145 146 147 148 149 150 151 152 153 154 155 156 157 158 159 160
 T   F   T   N   H   V   N   A   W   K   S   H   G   M   N   L
ACG TTC ACC AAT CAC GTG AAT GCA TGG AAA TCC CAC GGT ATG AAC CTA
TGC AAG TGG TTA GTG CAC TTA CGT ACC TTT AGG GAG CCA TAC TTG GAT
                    NsiI                                  StyI/
          A152C mutagenic primer site
161 162 163 164 165 166 167 168 169 170 171 172 173 174 175 176
 G   S   N   W   A   Y   Q   V   M   A   T   E   G   Y   Q   S
GGT TCT AAT TGG GCT TAT CAA GTA ATG GCG ACC GAA GGC TAC CAG AGC
CCA AGA TTA ACC CGA ATA GTT CAT TAC CGC TGG CTT CCG ATG GAC TCG
SecI/AvrII

177 178 179 180 181 182 183 184 185 STOP
 S   G   S   S   N   V   T   V   W
TCT GGT TCT TCC AAC GTT ACA GTG TGG TAA AGATCTTGAAGCTT
AGA CCA AGA AGG TTG CAA TGT CAC ACC ATT TCTAGAACTTCGAA
SacI                                  BglII   HindIII
```

FIGURE 7 CONT'D

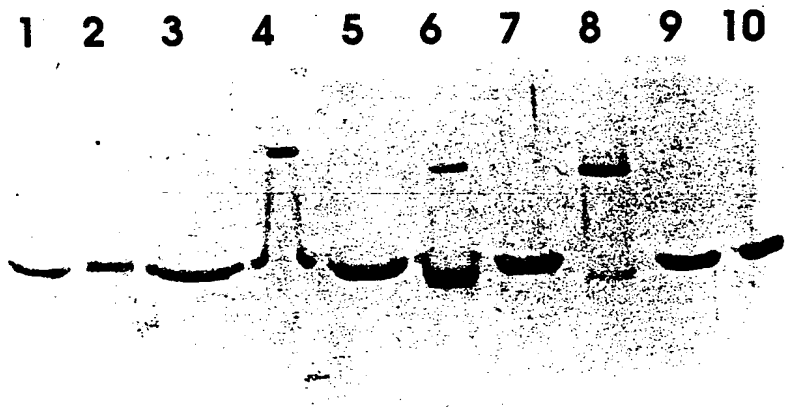


FIGURE 8

```
  1   2   3   4   5   6   7   8   9  10  11  12  13  14  15  16
  A   S   T   D   Y   W   Q   N   W   T   D   G   G   G   I   V
GCT AGC ACA GAT TAC TGG CAA AAC TGG ACA GAC GGT GGC GGT ATC GTT
CGA TCG TGT CTA ATG ACC GTT TTG ACC TGT CTG CCA CCG CCA TAG CAA
 NheI

 17  18  19  20  21  22  23  24  25  26  27  28  29  30  31  32
  N   A   V   N   G   S   G   G   N   Y   S   V   N   W   S   N
AAT GCC GTG AAC GGC TCC GGA GGC AAC TAC AGC GTG AAT TGG TCT AAT
TTA CGG CAC TTG CCG AGG CCT CCG TTG ATG TCG CAC TTA ACC AGA TTA
                   BspEI

 33  34  35  36  37  38  39  40  41  42  43  44  45  46  47  48
  T   G   N   F   V   V   G   K   G   W   T   T   G   S   P   F
ACT GGG AAC TTC GTA GTC GGA AAA GGT TGG ACG ACA GGA TCC CCG TTC
TGA CCC TTG AAG CAT CAG CCT TTT CCA ACC TGC TGT CCT AGG GGC AAG
                                               BamHI

 49  50  51  52  53  54  55  56  57  58  59  60  61  62  63  64
  R   T   I   N   Y   N   A   G   V   W   A   P   N   G   N   G
CGT ACG ATC AAC TAC AAC GCT GGC GTT TGG GCC CCG AAT GGT AAC GGT
GCA TGC TAG TTG ATG TTG CGA CCG CAA ACC CGG GGC TTA CCA TTG CCA
 SplI                                 ApaI

 65  66  67  68  69  70  71  72  73  74  75  76  77  78  79  80
  Y   L   T   L   Y   G   W   T   R   S   P   L   I   E   Y   Y
TAC CTG ACA CTG TAT GGC TGG ACG CGT TCG CCA CTG ATT GAA TAT TAC
ATG GAC TGT GAC ATA CCG ACC TGC GCA AGC GGT GAC TAA CTT ATA ATG
                       MluI                     SspI

 81  82  83  84  85  86  87  88  89  90  91  92  93  94  95  96
  V   V   D   S   W   G   T   Y   R   P   T   G   T   Y   K   G
GTT GTC GAC TCT TGG GGA ACG TAC CGT CCG ACT GGA ACC TAC AAA GGC
CAA CAG CTG AGA ACC CCT TGC ATG GCA GGC TGA CCT TGG ATG TTT CCG
     SalI

 97  98  99 100 101 102 103 104 105 106 107 108 109 110 111 112
  T   V   K   S   D   G   G   T   Y   D   I   Y   T   T   T   R
ACA GTC AAA AGC GAT GGT GGT ACC TAC GAC ATC TAC ACC ACC ACA AGA
TGT CAG TTT TCG CTA CCA CCA TGG ATG CTG TAG ATG TGG TGG TGT TCT
                       KpnI

113 114 115 116 117 118 119 120 121 122 123 124 125 126 127 128
  Y   N   A   P   S   I   D   G   D   R   T   T   F   T   Q   Y
TAC AAC GCA CCT TCC ATC GAT GGC GAT CGG ACC ACC TTT ACT CAG TAC
ATG TTG CGT GGA AGG TAG CTA CCG CTA GCC TGG TGG AAA TGA GTC ATG
                   ClaI    PvuI
```

FIGURE 9

```
129 130 131 132 133 134 135 136 137 138 139 140 141 142 143 144
 W   S   V   R   Q   S   K   R   P   T   G   S   N   A   T   I
TGG AGT GTT AGA CAA TCT AAG CGG CCG ACT GGT TCG AAC GCC ACC ATT
ACC TCA CAA TCT GTT AGA TTC GCC GGG TGA CCA AGC TTG CGG TGG TAA
                           EagI/EaeI    BstBI

145 146 147 148 149 150 151 152 153 154 155 156 157 158 159 160
 T   F   T   N   H   V   N   A   W   K   S   H   G   M   N   L
ACG TTC ACC AAT CAC GTG AAT GCA TGG AAA TCC CAC GGT ATG AAC CTA
TGC AAG TGG TTA GTG CAC TTA CGT ACC TTT AGG GAG CCA TAC TTG GAT
                       NsiI                                StyI/

161 162 163 164 165 166 167 168 169 170 171 172 173 174 175 176
 G   S   N   W   A   Y   Q   V   M   A   T   E   G   Y   Q   S
GGT TCT AAT TGG GCT TAT CAA GTA ATG GCG ACC GAA GGC TAC CAG AGC
CCA AGA TTA ACC CGA ATA GTT CAT TAC CGC TGG CTT CCG ATG GAC TCG
SecI/AvrII

177 178 179 180 181 182 183 184 185 STOP
 S   G   S   S   N   V   T   V   W
TCT GGT TCT TCC AAC GTT ACA GTG TGG TAA AGATCTTGAAGCTT
AGA CCA AGA AGG TTG CAA TGT CAC ACC ATT TCTAGAACTTCGAA
SacI                                   BglII   HindIII
```

FIGURE 9 CONT'D

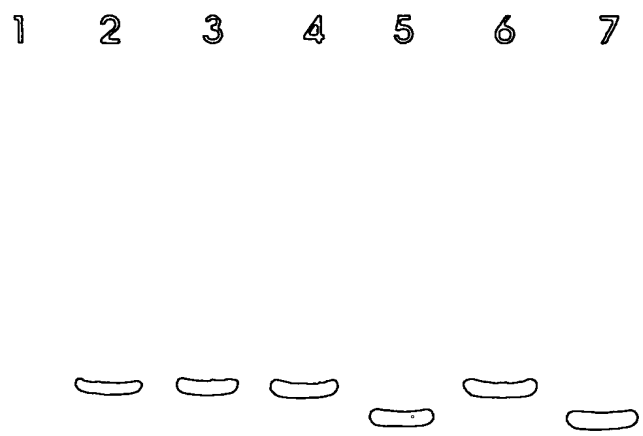


FIGURE 10

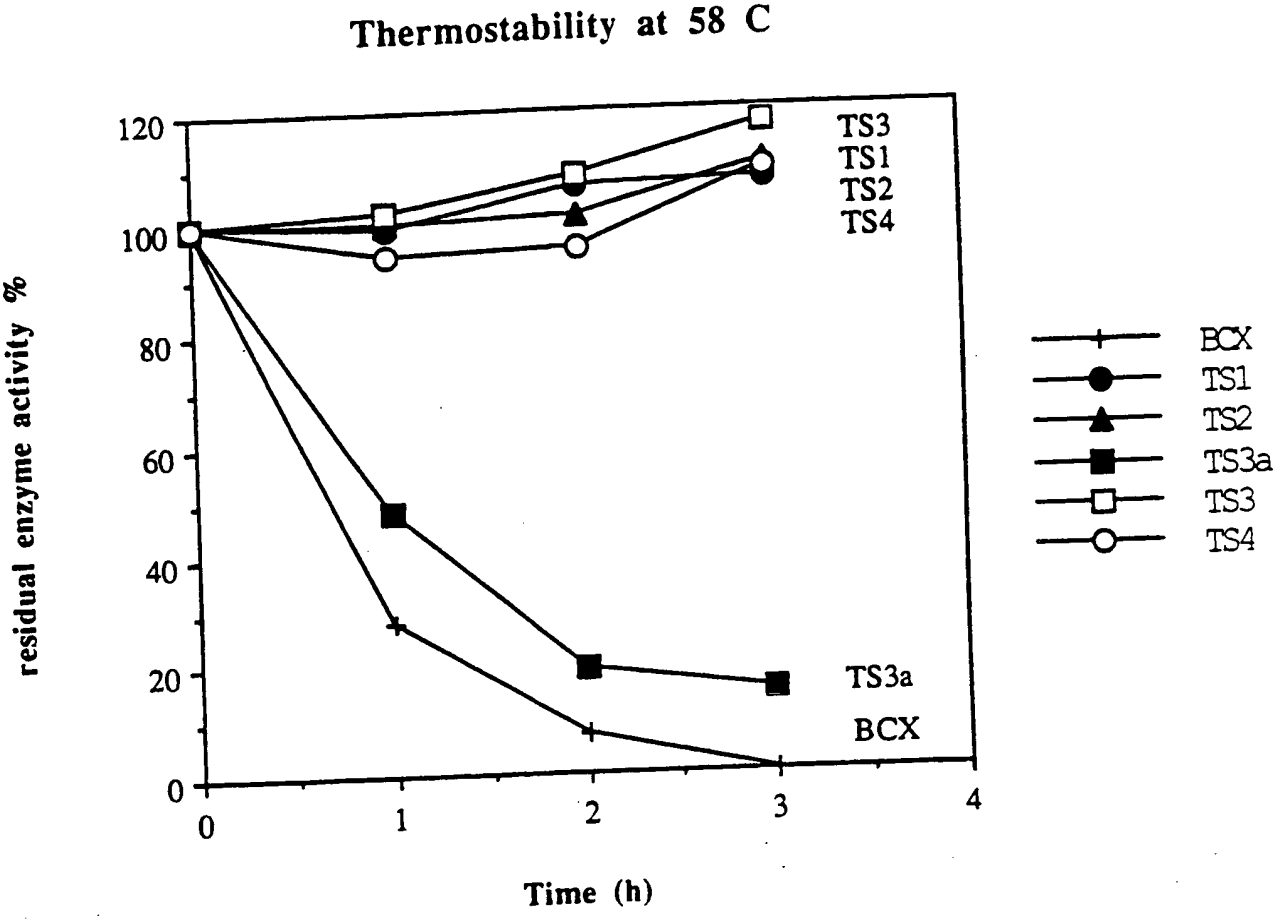


FIGURE 11

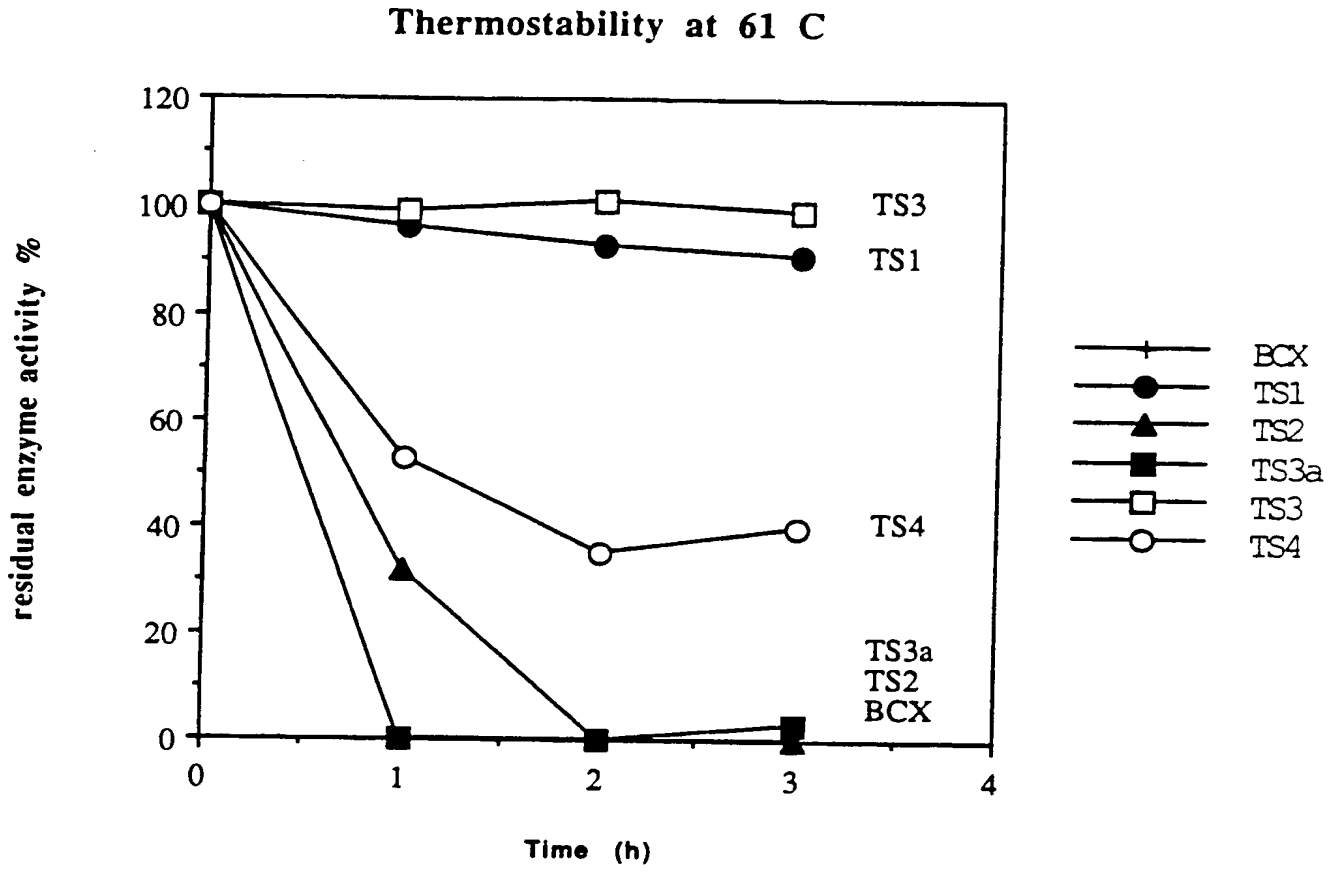


FIGURE 12

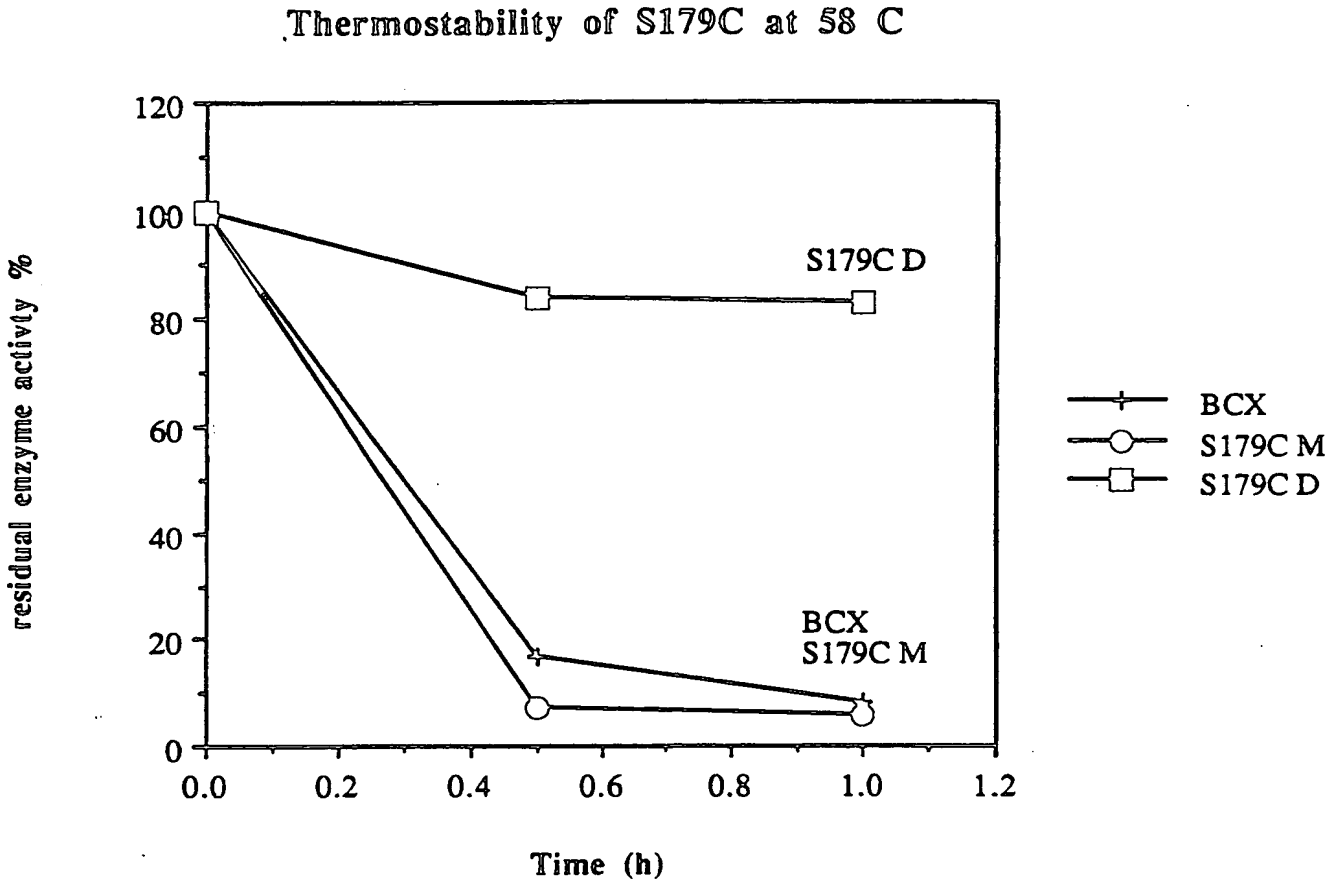


FIGURE 13

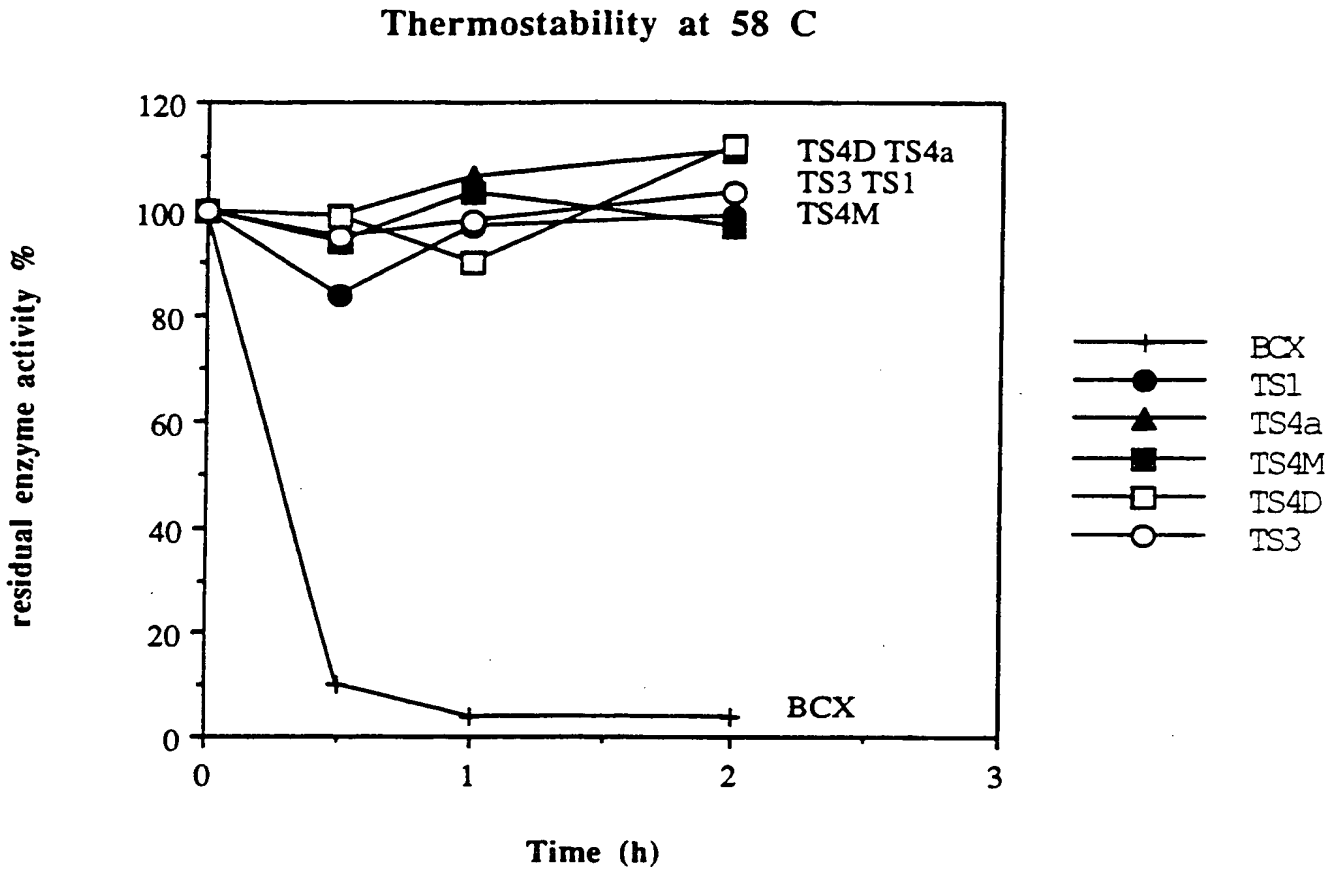


FIGURE 14

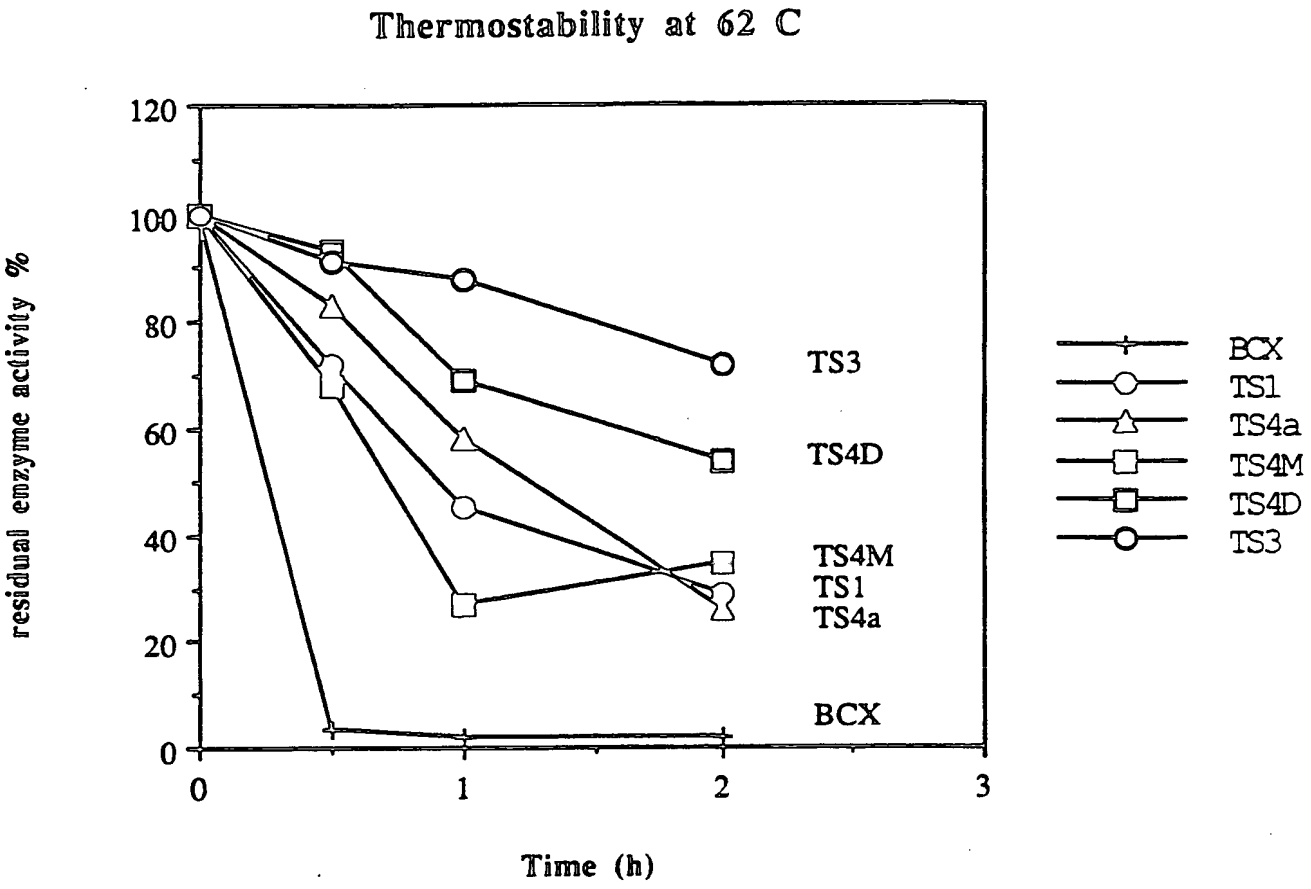
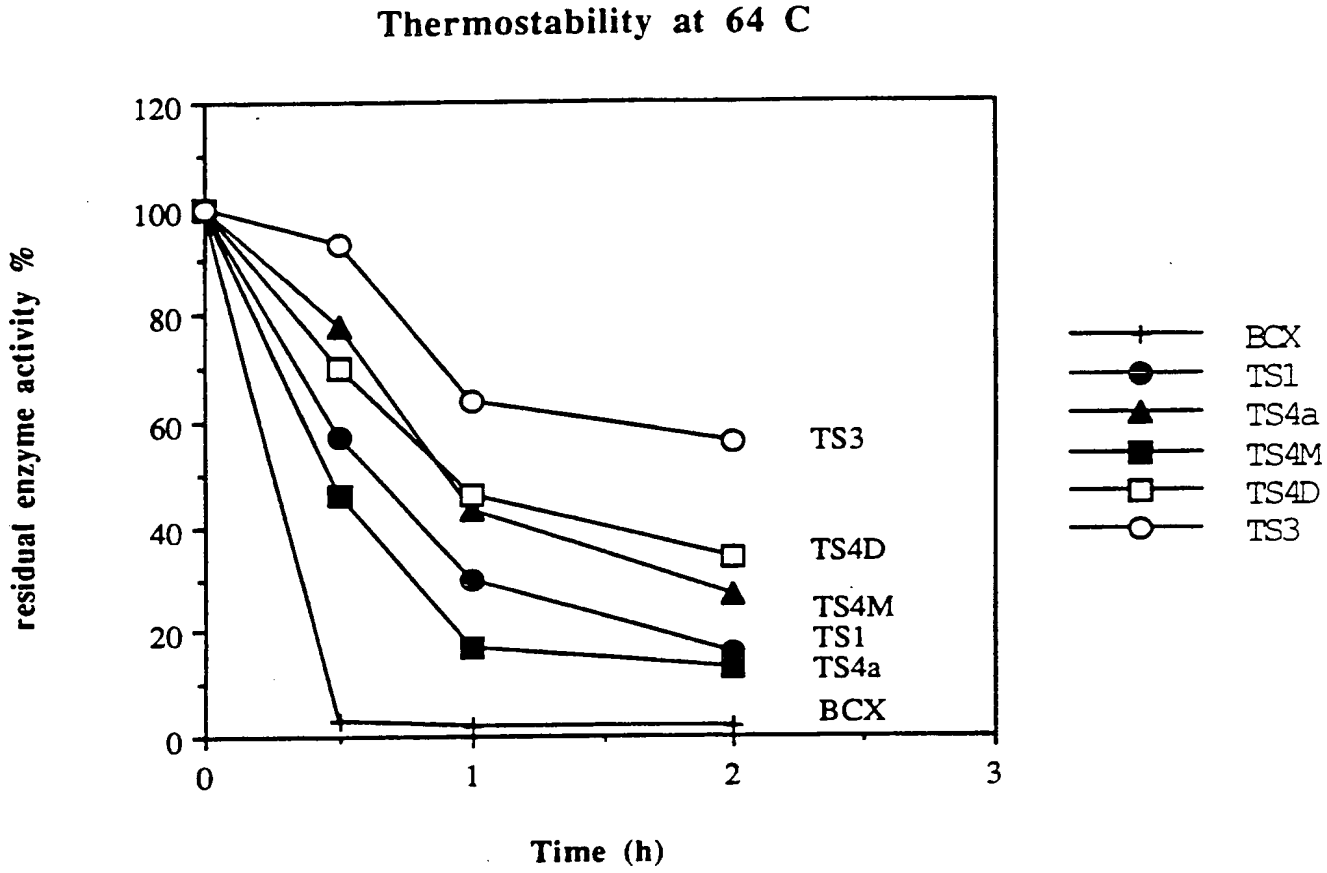


FIGURE 15



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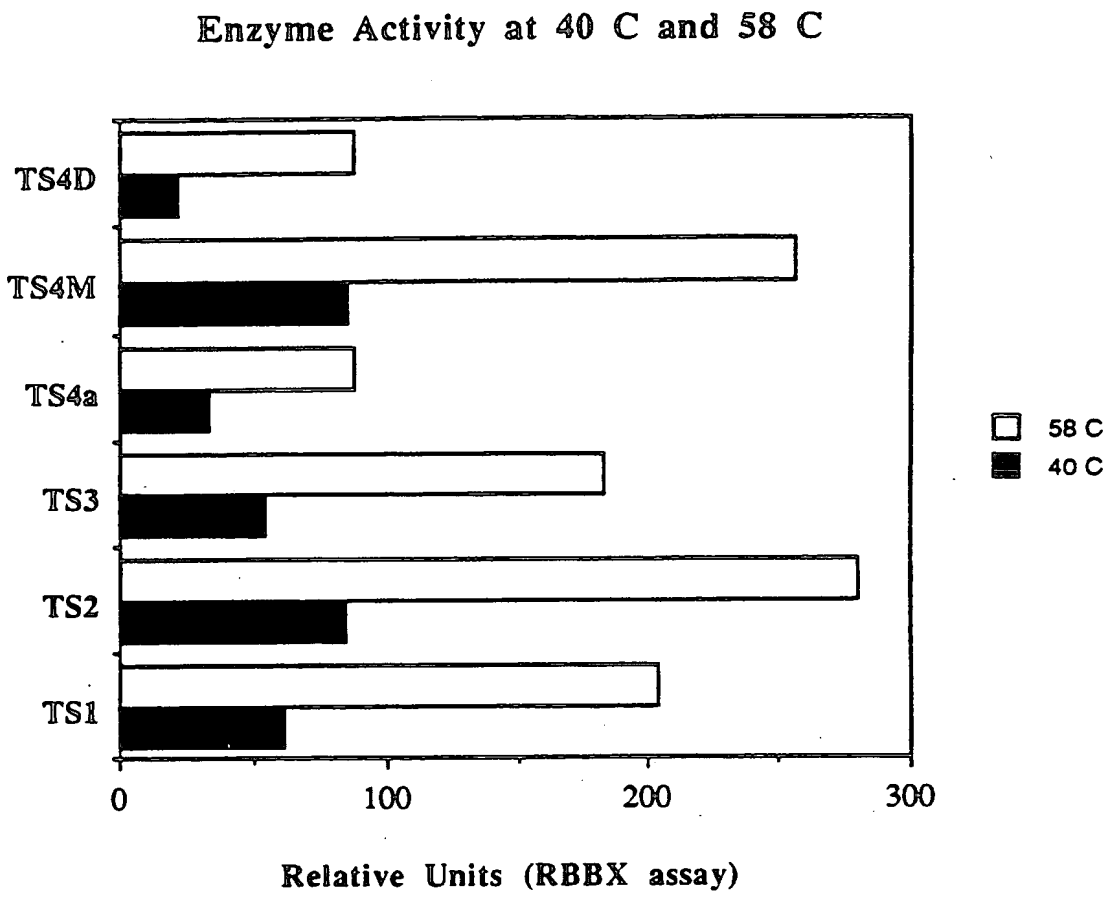

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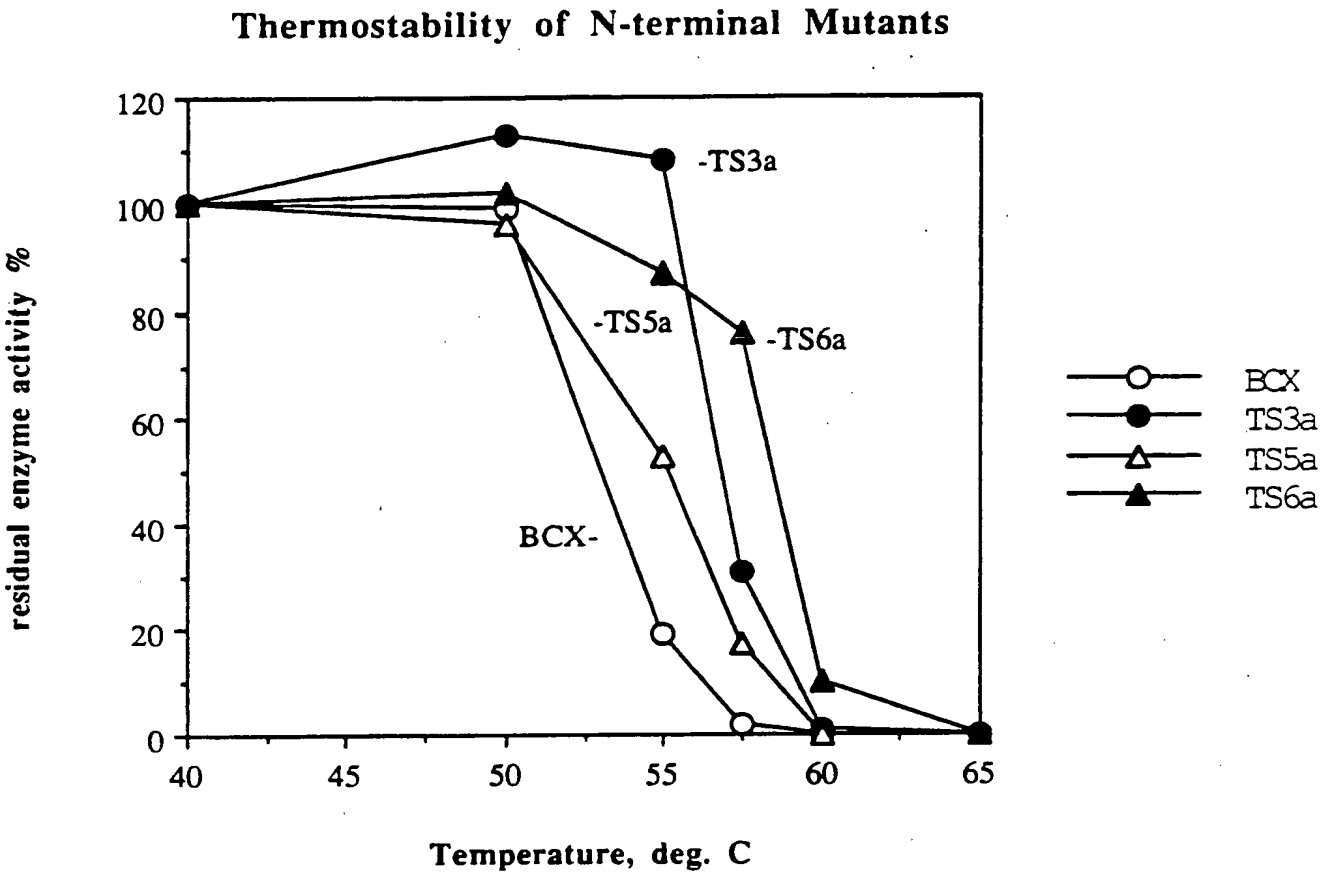

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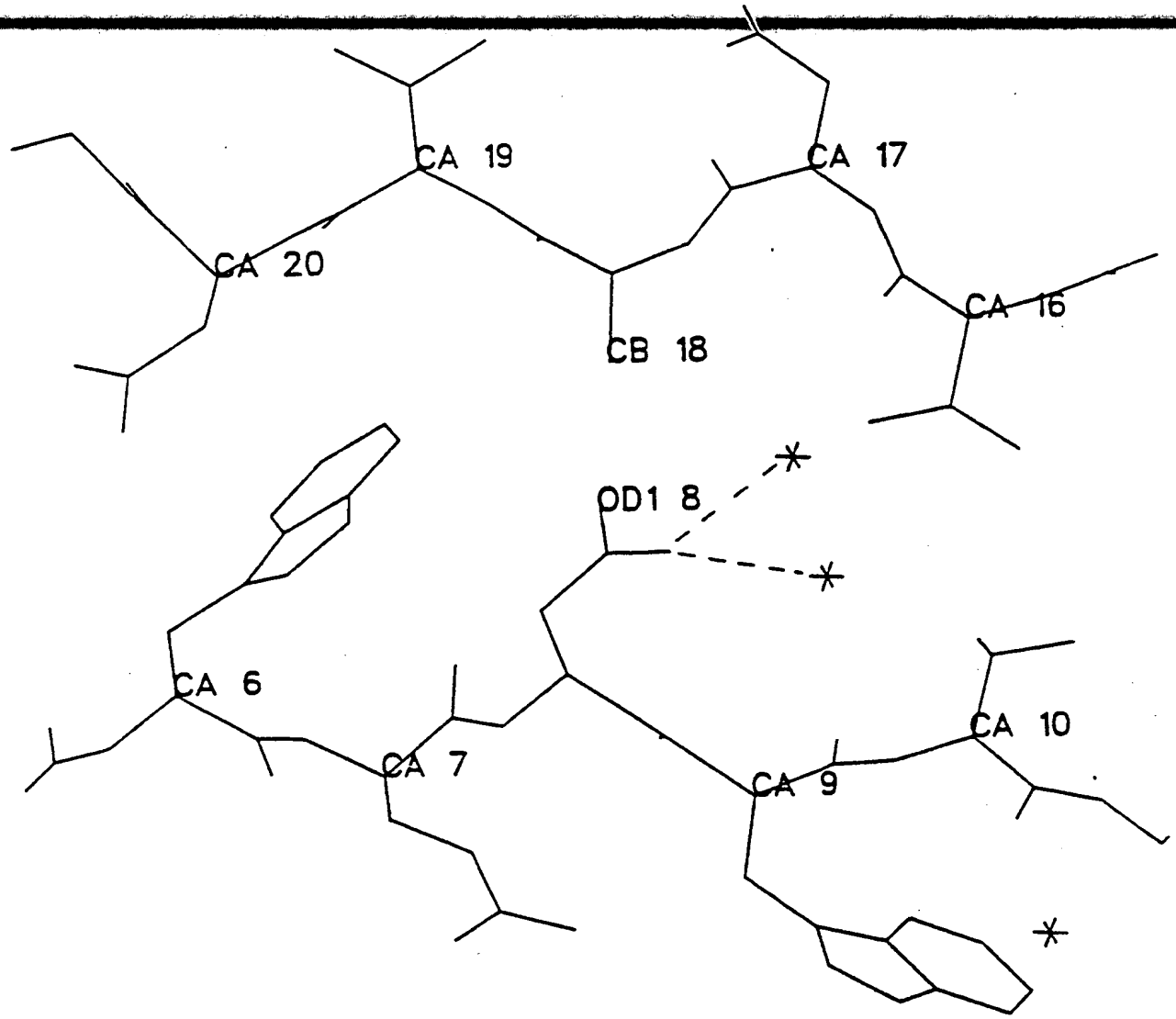

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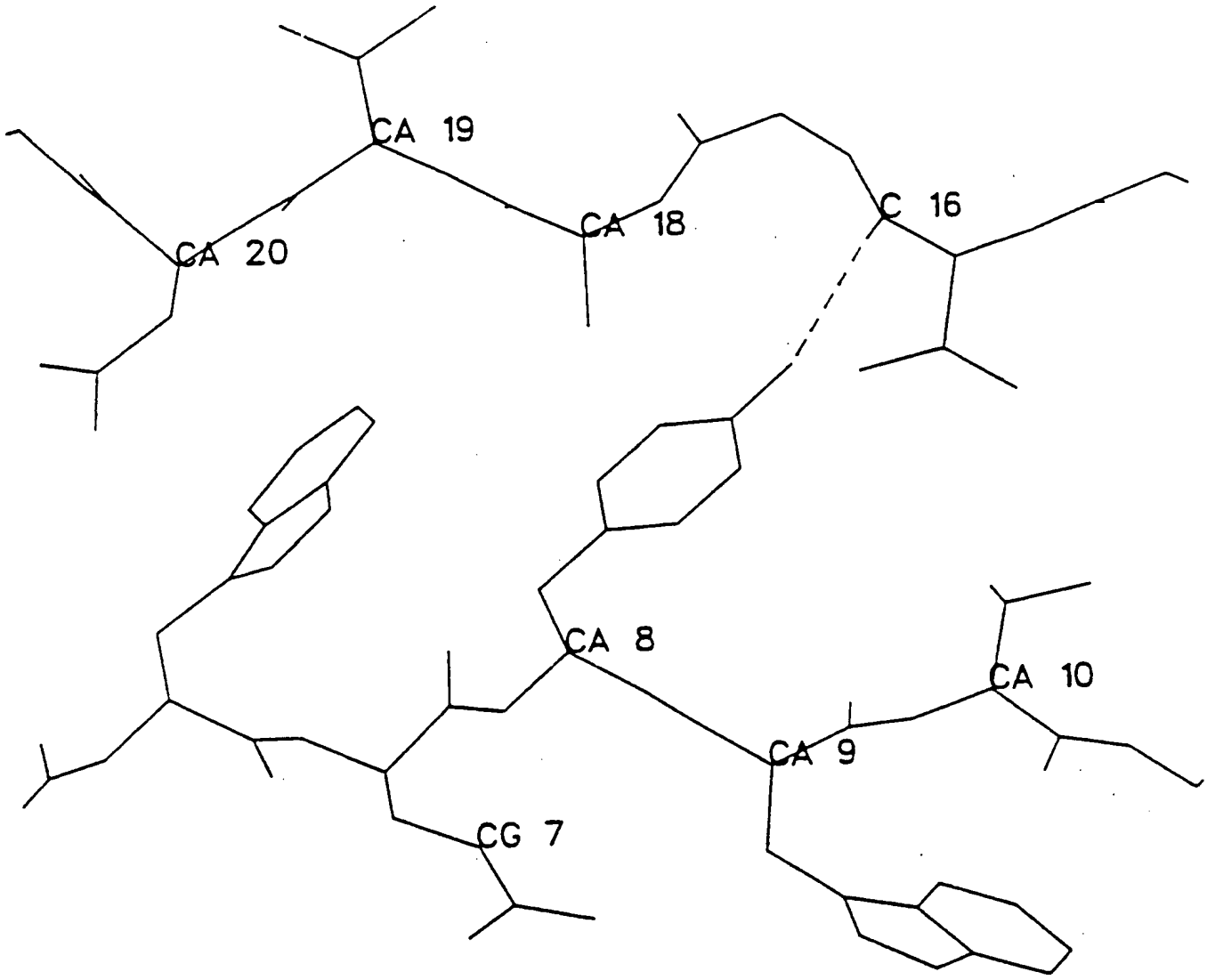

FIGURE 20

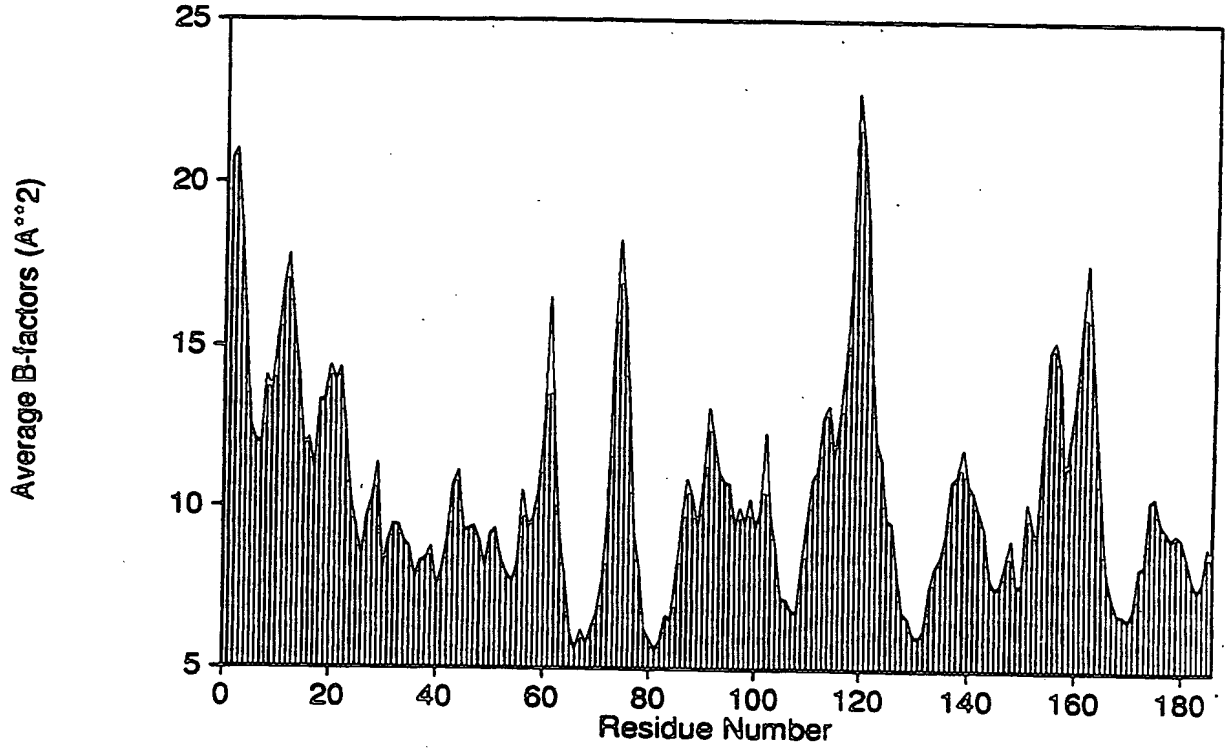

FIGURE 21

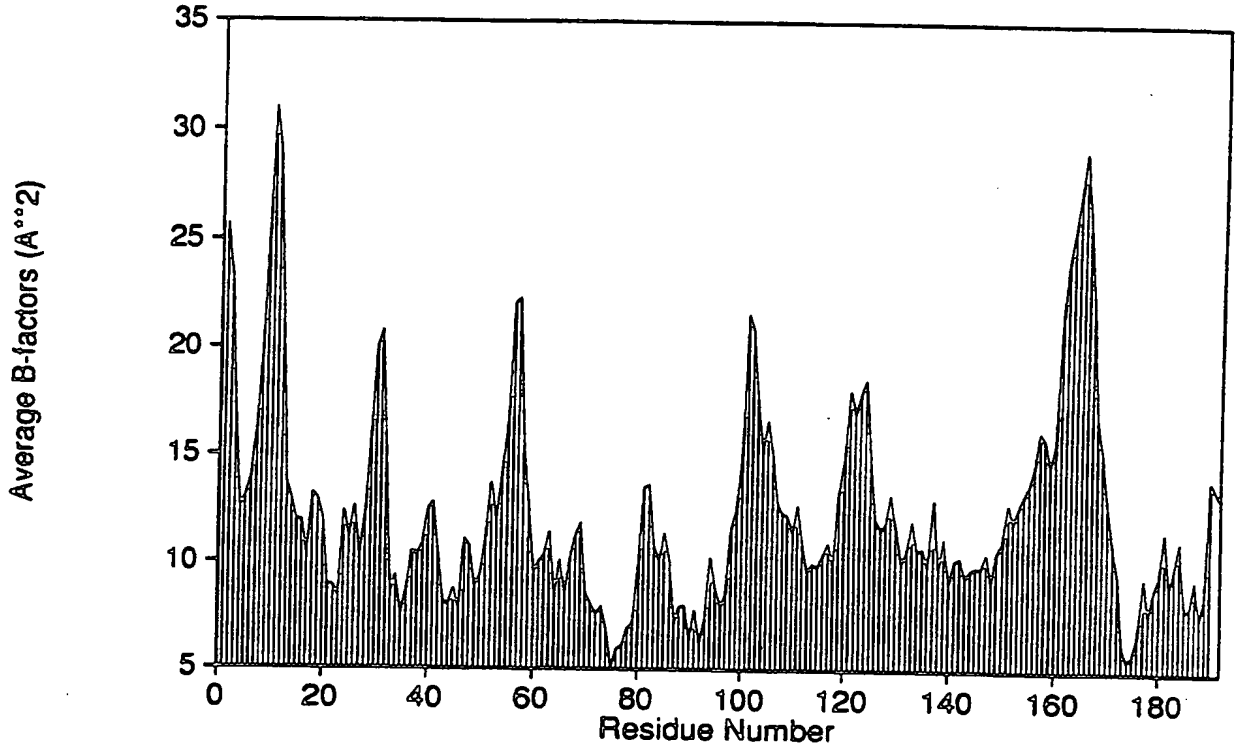

FIGURE 22

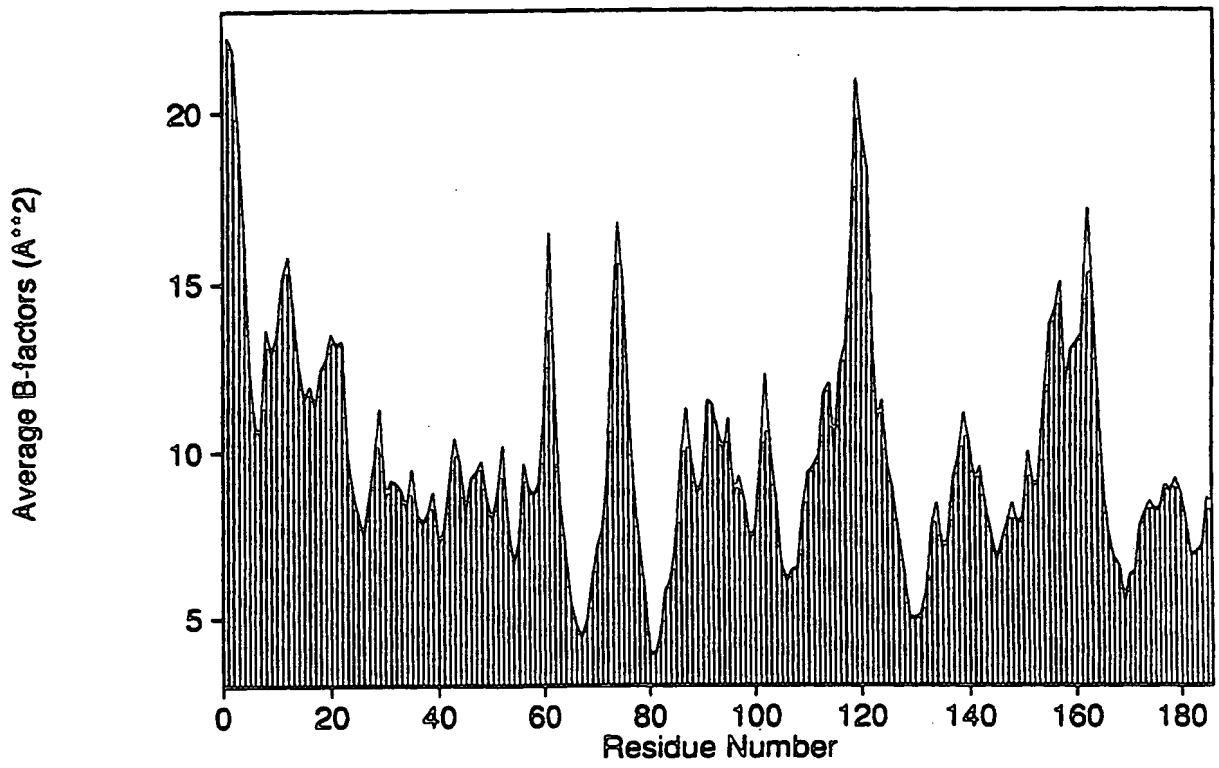

FIGURE 23

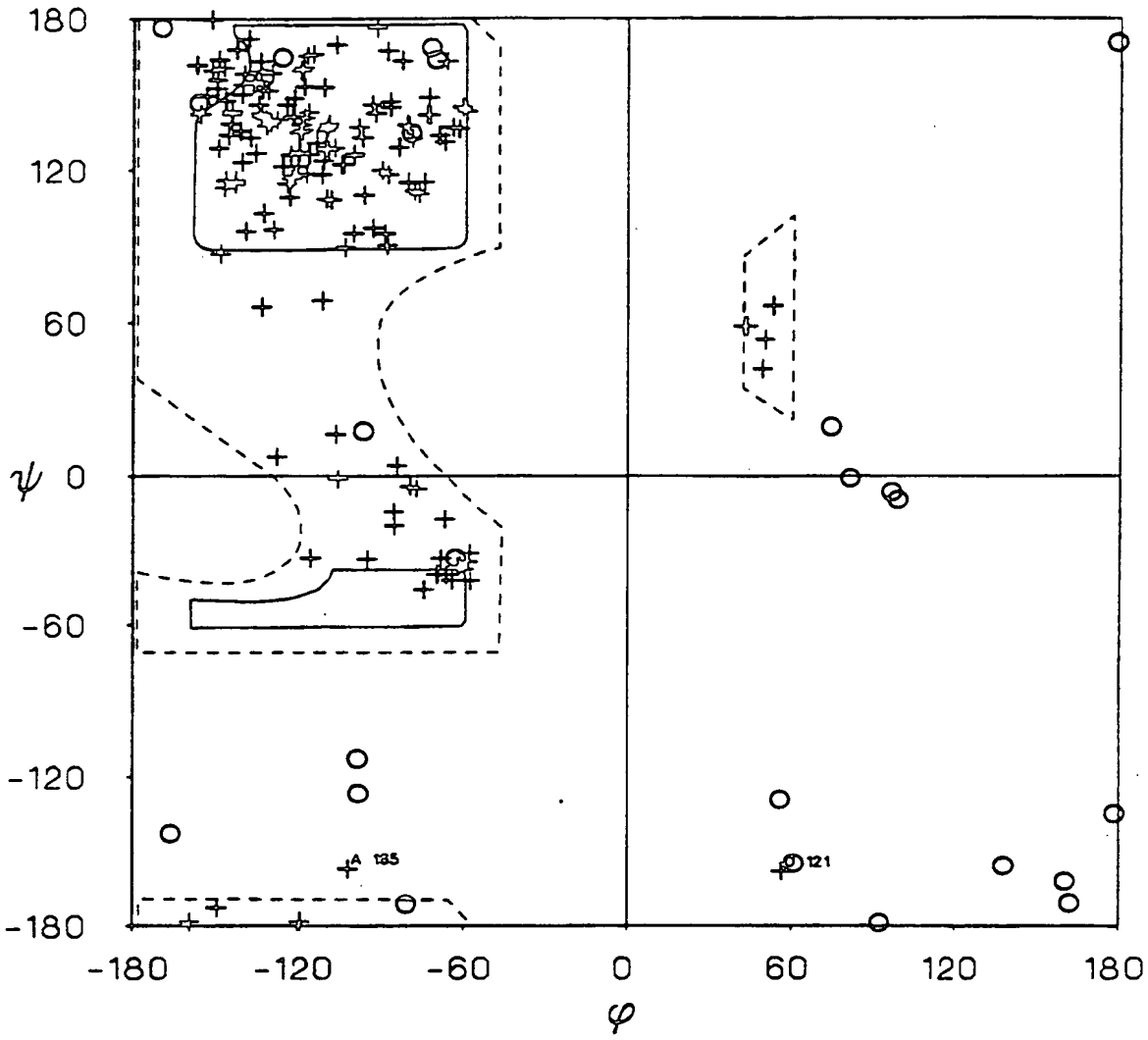

FIGURE 24

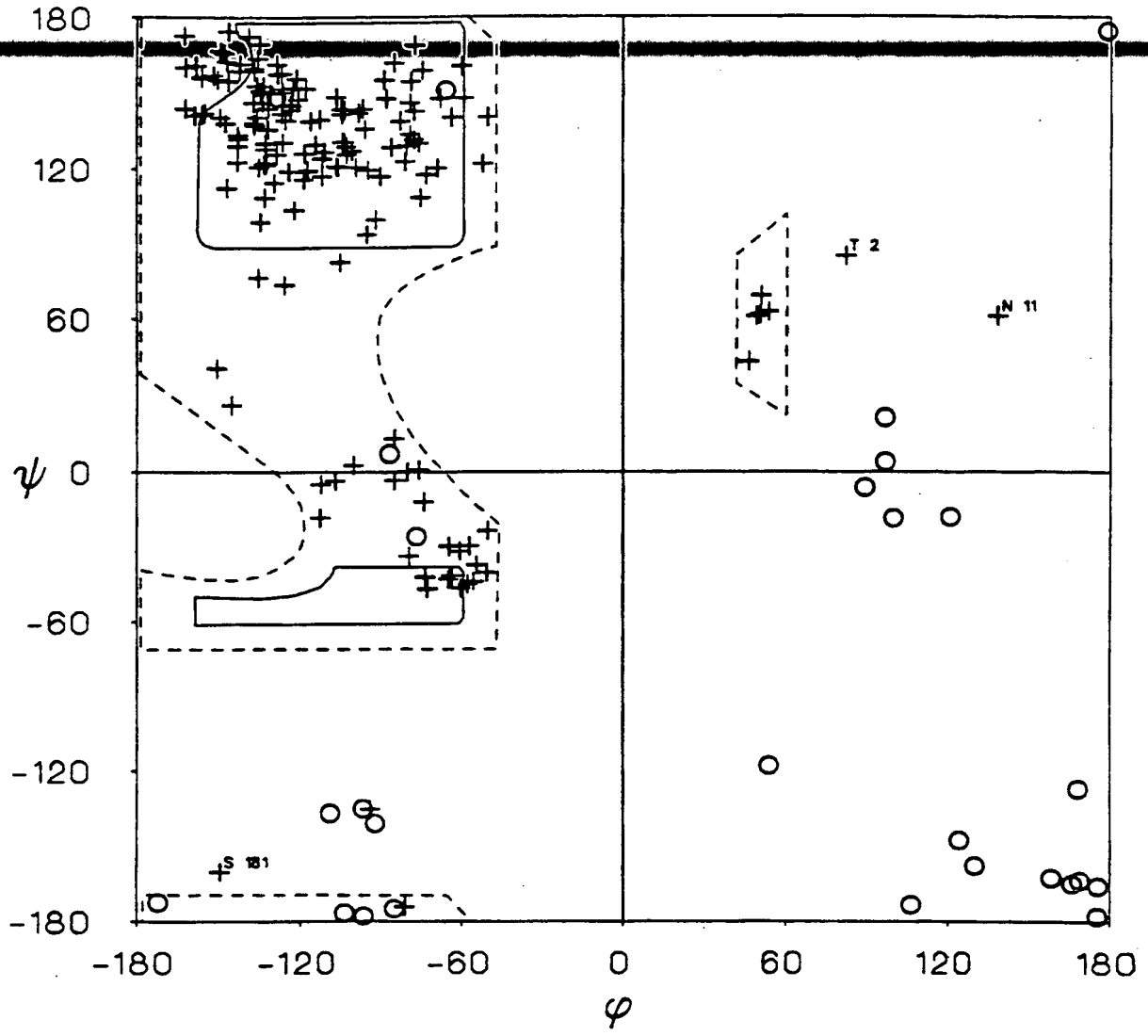

FIGURE 25

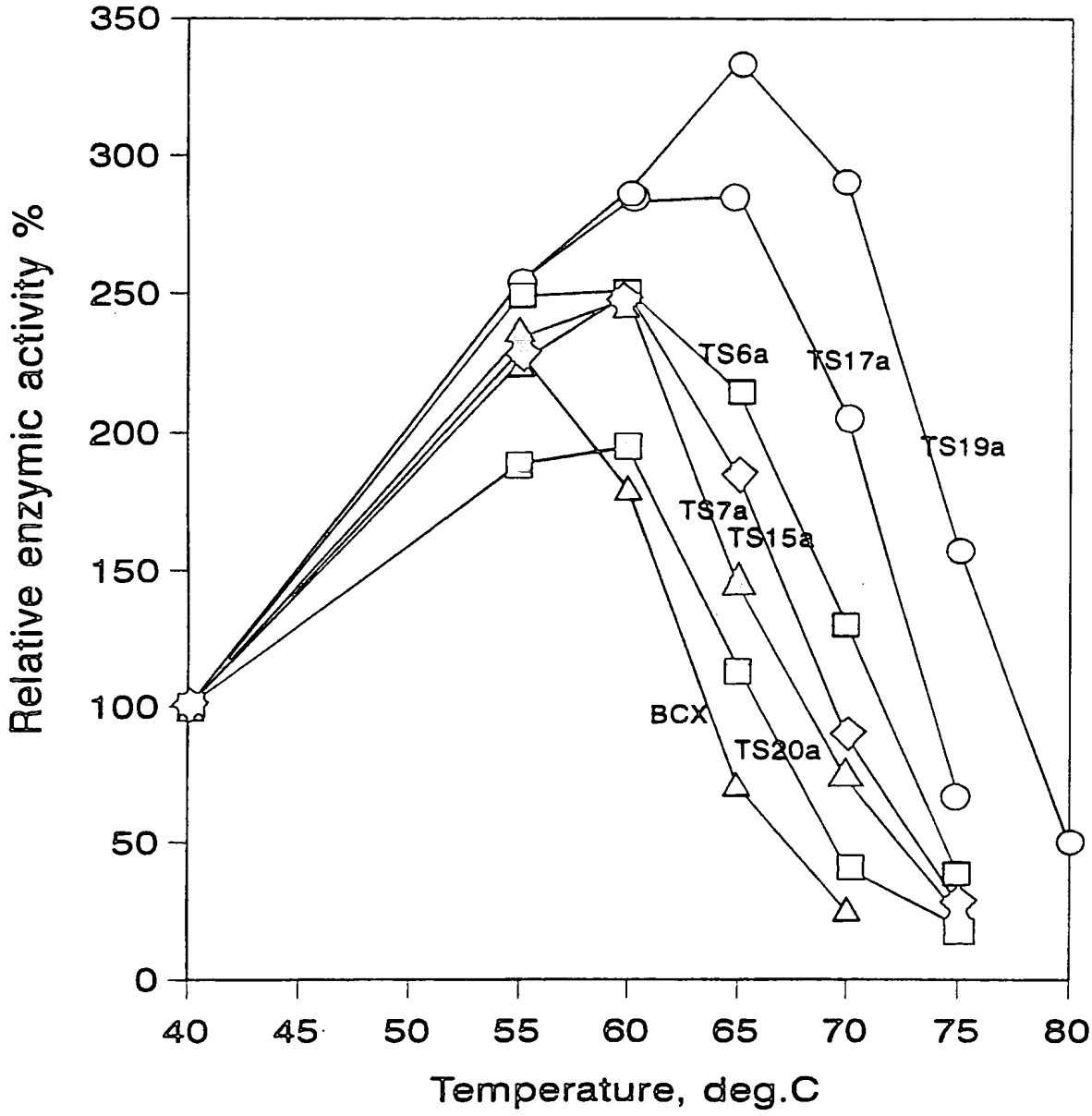

FIGURE 26

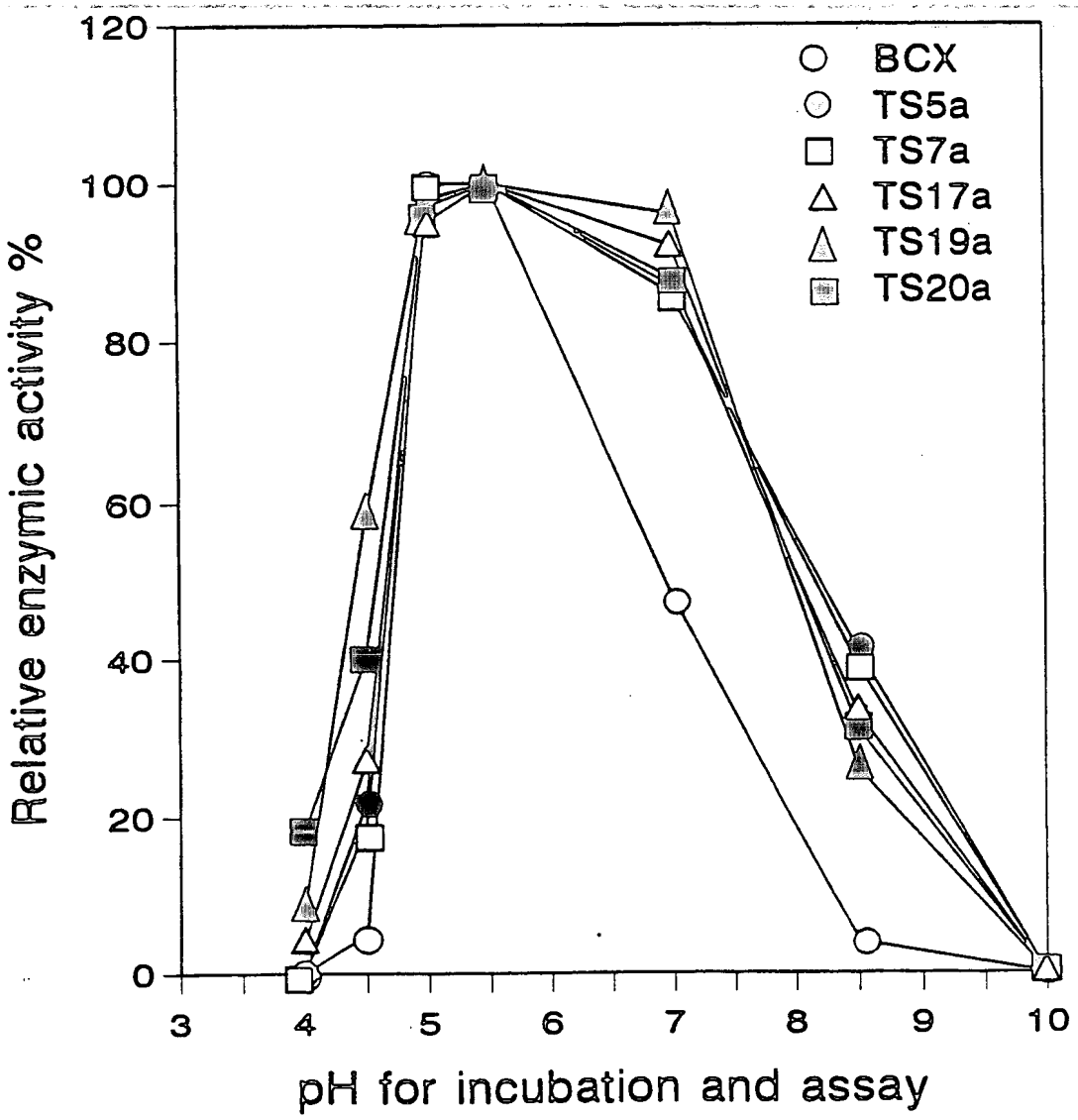

FIGURE 27

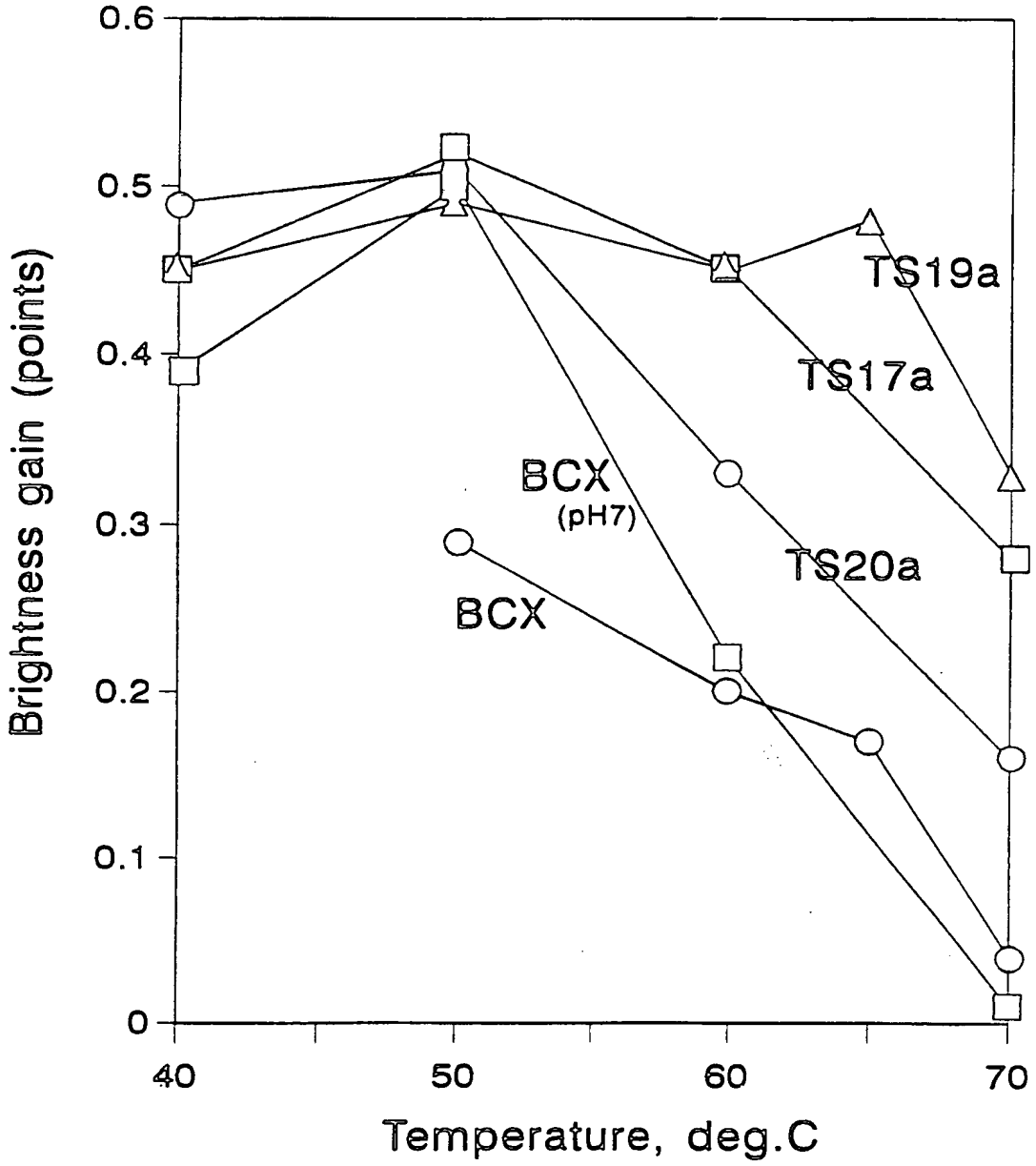

FIGURE 28